

THE AMERICAN NATURALIST

VOL. LI.

December, 1917

No. 612

THE GENESIS OF THE ORGANIZATION OF THE INSECT EGG. II

PROFESSOR ROBERT W. HEGNER,

ZOOLOGICAL LABORATORY, UNIVERSITY OF MICHIGAN

5. Interaction of Nucleoplasm and Cytoplasm

There are phenomena that occur during the growth period that suggest how masses of cytoplasm that are differentiated both morphologically and physiologically may arise in the cortical layer of the insect egg. It has been suggested that "most of the differentiations of the egg cytoplasm have arisen during the ovarian history of the egg and as a result of the interaction of nucleus and cytoplasm; . . .,"²⁷ and with this we fully agree, but our problem is to determine the nature of this interaction and in what ways it may take place.

During every mitosis there is a more or less thorough mixing that involves the chromatin as well as other nuclear constituents, since chromatin-diminution is a normal histological process. Interchanges between nucleus and cytoplasm, therefore, occur during the two multiplication periods that precede the formation of oocytes. Abundant opportunity is thus offered for factors in the chromosomes to exert an influence upon the cell as a whole. A similar and probably even greater discharge of chromatic

²⁷ Conklin, 1916, "Heredity and Environment," New York.

and other nuclear substances into the cytoplasm occurs during the maturation divisions of the egg, but this period may be neglected in this connection, since the organization with which we are concerned is already established before maturation takes place. Even when the nuclear membrane is intact, substances undoubtedly pass in and out of the nucleus much as they do through the cell membrane, and as in the latter, the nuclear membrane may change in permeability at different times, these changes being due to chemical processes taking place within the nucleus or in the cytoplasm. Such changes occur more often during periods of cell activity than at other times and thus we should expect pronounced interaction throughout the growth period of the oocytes.

Besides gradual, and for the most part invisible, interchanges of this sort there may be actual transference of visible masses of chromatin from the nucleus to the cytoplasm. These chromatin granules that escape into the cytoplasm have been called "chromidia" and are supposed to play a part in cytoplasmic differentiation.

A peculiar process of interchange by means of secondary nuclei is exhibited by certain insects, especially Hymenoptera.²⁸ This process has been studied most carefully in the carpenter ant, *Componotus herculeanus* var. *pennsylvanica* (Fig. 11). At an early stage in the growth of the oocyte small vesicles containing a few granules of chromatin appear near the oocyte nuclei. These "secondary nuclei" appear to arise as buds from the primary nucleus, but no one has yet actually observed their formation in this way. It has also been suggested that they may be epithelial cells that have invaded the oocyte, but this seems very improbable. The writer has reached the conclusion that they consist of nuclear materials that have been given off into the cytoplasm and have there become enclosed by membranes which give them a nuclear-like appearance. As the oocyte increases

²⁸ Blockmann, 1886, *Festsch. nat.-med. Verein zu Heidelberg*; Buchner, 1913, *Biol. Centrbl.*, Bd. 33; Hegner, 1915, *Journ. Morph.*, Vol. 26.

in size the secondary nuclei increase in number until they entirely surround the primary nucleus, forming several layers. When the oocyte has nearly reached its full growth they begin to migrate from the group near the anterior end of the oocyte and become scattered through-

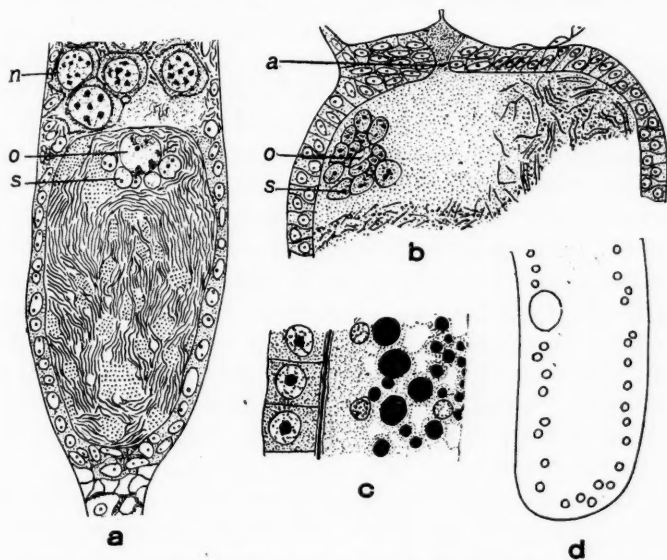


FIG. 11. Secondary nuclei in the oocytes of the carpenter ant (*a*, *b*, *c*) and the Hymenopterous gall-fly, *Rhodites ignota* (*d*). (Hegner, 1915.)

a. Oocyte (*o*) shortly after secondary nuclei (*s*) begin to appear. *n* = nurse cells.

b. Older oocyte showing oocyte nucleus (*o*) surrounded by secondary nuclei (*s*). *a* = connection between oocyte and nurse chamber.

c. Part of a still older oocyte showing follicular epithelium, yolk globules (black), and secondary nuclei.

d. Part of an oocyte of *Rhodites* showing primary nucleus (large circle) and secondary nuclei (small circles).

out the egg, forming a rather regular layer a short distance beneath the periphery. The further history of these bodies is not certain, but they undergo changes by which they lose their identity, since they can not be found in fully grown eggs. Their function is likewise problematical. They may take part in the formation of germ-line determinants which probably occur in the eggs of

this ant;²⁹ they may aid in changing the substances furnished by the nurse cells into material available for the embryo;³⁰ or they may have something to do with the formation of yolk.³¹ It is also possible that they may control differentiation in the peripheral layer of cytoplasm and thus provide a method of nuclear control of the organization of the egg. The last hypothesis may be objected to on the grounds that the secondary nuclei appear to be irregularly distributed and that they are known to occur in only a few species of insects.

Another possible way in which the initial organization of the insect egg may arise is through the activities of mitochondria. The rather constant presence of these bodies in the cytoplasm of almost all types of cells indicates that they may be of considerable importance in the process of differentiation. If they take part in the genesis of egg organization they then may play the rôle attributed to them by certain investigators of being the cytoplasmic bearers of hereditary factors corresponding in this respect to the nuclear bodies of similar function, the chromosomes.

The most striking differentiation in the cytoplasm of the insect egg is that which involves the germ-line determinants. As stated above, we do not know for certain in any case the origin of the peculiar cytoplasmic mass that contains these determinants, but a number of hypotheses have been suggested. In *Miastor*, for example, the following is offered to account for the appearance of the "pole-plasm" in the fully developed oocyte.³²

It may be distinguished from the rest of the egg contents by its position at the posterior end and because of its affinity for certain dyes. It appears shortly before the maturation division is initiated, but no transition stages have been discovered—it has been either present or entirely absent in the preparations thus far studied. If we consider the history of this substance from the formation of the primordial germ

²⁹ Hegner, 1914, *Journ. Morph.*, Vol. 26.

³⁰ Marshall, 1907, *Zeit. wiss. Zool.*, Bd. 86.

³¹ Loyez, 1908, *C. R. Assoc. Anat.*, 10 Reunion, Marseille.

³² Hegner, 1914, "Germ-Cell Cycle in Animals," New York.

cell to the growth period of the oocytes produced by this primordial germ cell, we may conclude that at the time the multiplication period ends the pole-plasm has become equally distributed among the sixty-four oogonia. Then ensues the growth period during which the pole-plasm can not be distinguished. Later, however, just before maturation, pole-plasm substance reappears which is equal in amount to that contained in the primordial germ cell of the preceding generation or to that contained in all of the sixty-four oogonia which descended from that primordial germ cell. That is, the pole-plasm of the oocyte under discussion has in some way increased until its mass is sixty-four times as great as that of the oogonium before the growth period began. How this increase has taken place can only be conjectured. The pole-plasm in the oogonium may have produced new material of its own kind either by the division of its constituent particles or by the influence of its presence.

The influence of a specialized mass of cytoplasm upon the chromatin is very well illustrated by the inhibition of chromatin-diminution in *Miastor* and *Ascaris*. In *Miastor* nuclear division is normal until at the four-cell stage one nucleus reaches the pole-plasm at the posterior end (Fig. 12, *a*, IV.). During the succeeding mitosis this nucleus, which is apparently under the control of the pole-plasm, does not undergo chromatin-diminution, whereas the other three do. One of the daughter nuclei resulting from the division of this undiminished nucleus remains entirely within the pole-plasm and is cut off from the rest of the egg with this specialized mass of cytoplasm as the primordial germ cell (Fig. 12, *b*). This nucleus always retains the full amount of chromatin; but its sister nucleus, which remains in the egg and is thus separated from the direct influence of the pole-plasm, undergoes diminution at the next mitosis.

A similar segregation of specialized cytoplasm in the primordial germ cells occurs also in certain other insects and in copepods, but no diminution process has yet been discovered in them. In *Ascaris*, where chromatin-diminution was first reported,³³ there is evidently a segregation of germinal cytoplasm at each cleavage division up to the sixteen-cell stage, when it is all confined in one cell, the

³³ Boveri, 1887, *Anat. Anz.*, Bd. 2.

primordial germ cell. This cytoplasm, which is not visibly different from the rest, as in *Miastor*, appears to inhibit diminution in every nucleus that comes within its

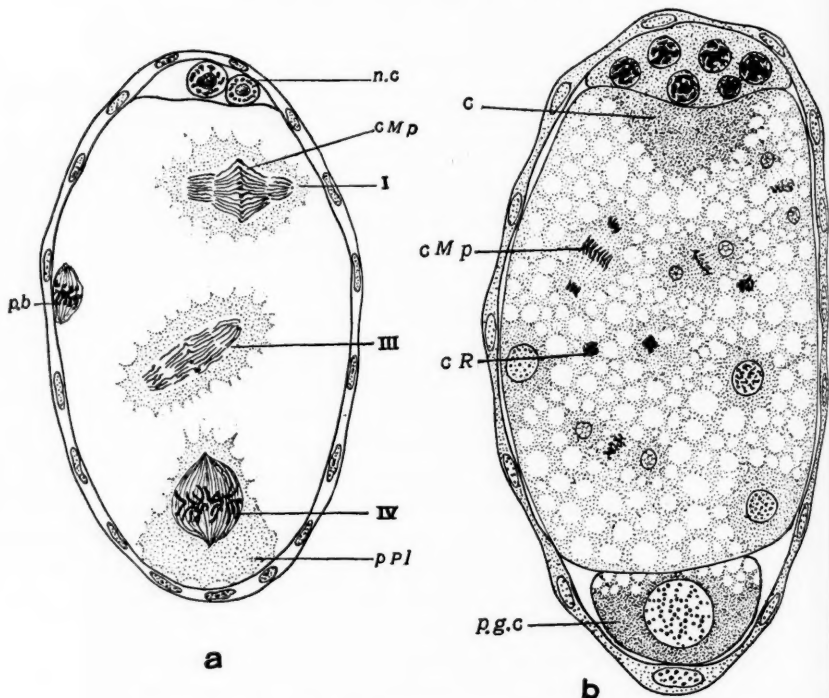


FIG. 12. *a*. Longitudinal section through an egg of *Miastor* showing chromatin-diminution in nuclei I. and III. but not in nucleus IV. which has come under the influence of the pole-plasm (*pPl*). Nucleus II. does not appear in this section. *cMp* = chromatin that is cast off into cytoplasm. *nc* = nurse cells. *pb* = polar body. (Kahle, 1908.)

b. Longitudinal section through an egg of *Miastor*, showing primordial germ cell (*p.g.c.*), nuclei undergoing chromatin-diminution (*cMp*), and the remains of chromatin cast out into the cytoplasm (*cR*), *c* = cytoplasm elaborated by nurse cells above. (Hegner, 1914.)

immediate influence as indicated by experimental studies on dispermic and centrifuged eggs.³⁴ In this respect it resembles the pole-plasm of *Miastor*.

³⁴ Boveri, 1910, *Arch. Ent.-mech.*, Bd. 30; Boveri, 1910, *Festschr. R. Hertwig*, Bd. 3.

6. Mendelian Factors and Cytoplasmic Organization

The central biological problem of the present time is the method of evolution, and a knowledge of the mechanism of heredity has long been recognized as necessary for its solution. The results derived from breeding experiments with the fruit fly, *Drosophila ampelophila*, have dominated the field of genetics for the past five years, but although of very great interest and importance, their evolutionary significance is not yet certain. To be of primary value from this viewpoint it is necessary to prove that new species may arise by means of Mendelian characters (mutations) such as white eye, miniature wing, club wing, etc. Since no one has ever been able to define satisfactorily what a species really is and hence what characters should be considered of specific value, this is a difficult problem.

The definitions given by two of our foremost authorities, one a systematist and the other a geneticist, are as follows: The systematist writes:³⁵

Forms of animals which present distinct assemblages of characters, in form, color and arrangement of parts under natural conditions, which are recognizable from descriptions and figures, should receive distinctive names and be catalogued, provided, of course, that the assemblage of characters includes all ontogenetic changes. If, in the examination of abundant material from different natural environments, we find these characters fairly constant, the forms may properly be called species; if not, varieties or races.

The geneticist writes:³⁶

Species may thus be distinguished by peculiarities of form, of number, of geometrical arrangement, of chemical constitution and properties, of sexual differentiation, of development and of many other properties. In any one or in several of these features together, species may be found distinguished from other species.

The mutations that have appeared in *Drosophila* do not become recognizable until a late stage in the life history of the individual, and are about the last characters

³⁵ Williston, 1908, AMER. NAT., Vol. 42.

³⁶ Bateson, 1913, "Problems of Genetics."

to appear in the individual development. They for the most part affect the size and shape of the wings, the size, shape and color of the eyes, and the color of the body.

If a systematist were asked whether these new races of *Drosophila* are comparable to wild species, he would not hesitate for a moment. He would call them all one species. If he were asked why, he would say, I think, "These races differ only in one or two striking points, while in a hundred other respects they are identical even to the minutest details." He would add, that as large a group of wild species of flies would show on the whole the reverse relations, viz., they would differ in nearly every detail and be identical in only a few points.³⁷

This point of view seems justified, since the foremost dipterologist in this country, a man who has named over one thousand species and genera, mostly of flies, says regarding the results of certain experiments carried on with *Drosophila* by one of his colleagues.³⁸

But I think it is absolutely certain—and I speak as an entomologist fairly familiar with flies—that it would be impossible to produce species of his sports even though they were bred for a thousand years.³⁹

In talking over this species question with one who has had considerable experience in systematic work⁴⁰ it became clear that although as a rule only a few of the more conspicuously contrasting characters are selected for descriptive purposes, as a matter of fact the individuals of different species are often different in practically every morphological characteristic. One who is very familiar with these species will realize these differences at once, although many of them are of such a nature that they can not be described so that any one else will recognize them. There seems to be no difficulty, however, in finding numerous describable contrasting characters in *Drosophila*, since at least fifty-nine are included in the descriptions of two recently named species⁴¹ that were selected

³⁷ Morgan, 1916, "Critique of the Theory of Evolution."

³⁸ Dr. F. E. Lutz.

³⁹ Williston, 1908, AMER. NAT., Vol. 42.

⁴⁰ Dr. Alexander G. Ruthven.

⁴¹ Sturtevant, 1916, *Annals Ent. Soc. Amer.*, Vol. 9.

at random, *D. superba* and *D. projectans*, and these characters relate to almost every part of the body. Many other differences would probably also be found between the physiological processes and general activities of the adults and between the morphological and physiological characteristics of the embryos, larvæ and pupæ of the two species if they were compared from these standpoints. It has been shown that the factor for the character club wing affects not only the character that gives this mutation its name, but also other characters, for example the presence or absence of a pair of spines on the sides of the thorax, these being always absent when the factor for club wing is present.⁴² It is possible that the combination of a number of such factors as that for club wing would ultimately satisfy the requirements of systematic entomologists and that new species could then be made up in the laboratory. Such mutations might therefore be of evolutionary value. If, however, these mutations fail to furnish characters of specific rank, or characters that may lead to the formation of new species, we must conclude that they are not of evolutionary significance, and look elsewhere for the factors that are responsible for specific characters and that may undergo changes which lead to transmutation.

Factors of this sort may lie in the chromosomes or in the cytoplasm, but they are probably the results of interaction between chromosomes and cytoplasm. As pointed out above, interaction of this sort has abundant opportunity to operate during the germ-cell cycle. The cytoplasmic differentiations resulting from the metabolic processes that culminate in the formation of an egg ready to undergo maturation are very striking in the case of insects, as indicated by observations and experiments on the eggs of chrysomelid beetles, and there seems to be no valid reason why the eggs of these beetles are different in their type and complexity of organization, both morphological and physiological, from those of *Drosophila*; for while we do

⁴² Morgan, Sturtevant, Muller, and Bridges, 1915, "Mechanism of Mendelian Heredity."

not know much about the growth of the egg and embryological development of this genus of flies, we do know that these processes in certain other flies resemble those of beetles.

If the Mendelian factors are located in the chromosomes, it is evident that they may exert an influence upon the entire contents of the egg, (1) during the mitotic divisions of the oogonia, (2) during the so-called resting stages of the oogonia, and (3) during the growth of the oocytes. It is also clear that all of the factors carried by the chromosomes have an equal opportunity to interact with the cytoplasm and not alone those that remain within the egg after the elimination of chromosomes during maturation. The adult, however, that develops from the egg, whether fertilized or unfertilized, exhibits only those detailed characteristics whose genetic factors are supposed to be located in the chromosomes remaining in the egg after maturation, or in those that are brought in by the sperm. This seems to indicate that none of these factors has any permanent influence upon the egg organization during the growth of the oocyte and until maturation is completed.

It seems impossible to ignore the chromosomes or even to locate the principal factors of heredity in any other cell bodies. It may therefore be necessary to reconstruct our ideas of chromosome architecture and thereby abandon the theory that these bodies consist of a linear series of factorial determiners for certain ferments and of nothing else. It may be possible to separate our hypothetical factors into two groups, (1) those responsible for such characteristics as the polarity, bilaterality and "pattern" of the egg, and (2) those that control mutations that appear at a late period in the life history like those that are so abundant in *Drosophila*. Perhaps the latter may be anchored to the chromosomes as has recently been suggested;⁴³ the main portion of the chromosomes might then represent the foundation for the factors responsible for the organization of the egg and the attached masses of

⁴³ Goldschmidt, 1917, *Genetics*, Vol. 2.

ferments might constitute the factors responsible for the modification of embryonic, larval and adult characters—factors such as have been employed for experimental breeding purposes by most geneticists. According to this hypothesis it would probably be necessary to consider the main portions of each chromosome as sufficient for the production of an entire organism. The fact that the group of factors carried by any one chromosome in *Drosophila* controls characters that are not restricted to any definite part of the body gives weight to this assumption.

Most geneticists are accustomed to deal with adult characters only, and on this account pay very little or no attention to the eggs, embryos and larvæ of the species they are experimenting with. But the eggs, embryos and larvæ contain all the factors for these adult characters, both those that are realized and those that are inhibited either by internal or external causes, and they may exhibit characters that make it possible to separate different lines although the adults may be indistinguishable. Furthermore, taxonomists have long recognized the value of embryonic characters as an aid in determining species.

We should always be careful to distinguish between the parts of the egg that are of hereditary significance and those that are not. Thus the shell or chorion of the silk-worm egg has been discussed under the heading of "cytoplasmic inheritance," whereas it is not a vital part of the egg, but, being secreted by the epithelium of the ovarian tube, is a well-defined characteristic of the adult female and its coloration, which follows the laws of Mendelian inheritance,⁴⁴ is controlled by maternal factors.

Such fundamental characteristics as polarity, symmetry, and pattern, which are so clearly exhibited by the eggs of insects and certain other animals, are much more difficult to study than adult characters and are probably not so easily modified. If any or all of them are carried over from one generation to another in the cytoplasm we have then a real instance of cytoplasmic inheritance. Even if this is the case the chromosomes doubtless exert

⁴⁴ Toyama, 1913, *Journ. of Genetics*, Vol. 2.

an influence upon the cytoplasm during the oogonial and growth periods of the egg, and a study of the genesis of cytoplasmic organization may lead to data that will help us solve this difficult problem.

If the polarity of the oocyte when recognizable is not inherited, *i. e.*, if it is not transmitted to the primordial germ cells by the egg, and retained by the oogonia, it must arise *de novo* just before or during the growth period. One observer⁴⁵ has found that in certain beetles the position of the spindle remains, resulting from the differential divisions that precede the formation of the oocyte, indicates the polarity of the ultimate organism, but he does not tell us how this "polarité predifferentielle" is brought about. In all insects the end of the egg directed toward the head of the mother becomes the anterior end of the offspring. This is also the pole of the egg lying next to the nurse cells or that is closest to the nurse-cell chamber. This relation between oocyte and nurse-cells may be the determining factor in the polarity of these eggs and, if so, would indicate that polarity here is due to environment. How this relation could influence the polarity may be explained by means of axial gradients of metabolism, such gradients in this case being produced by greater external stimulation at the end near the nurse-cell chamber where nutritive substances are elaborated and added to the oocyte. By this theory of metabolic gradients, differentiation along an antero-posterior axis can be accounted for and further differentiations of a morphological and physiological nature would result from "chemical transportative correlation between the different parts."⁴⁶

We should not lose sight of the fact, however, that these hypothecated physiological activities require protoplasm as a material basis and that their results depend upon the character of this protoplasm. If polarity is established at the stage suggested above, it follows a long series of nucleo-cytoplasmic interactions which have no doubt resulted in the differentiation and localization of numerous

⁴⁵ Govaerts, 1913, *Arch. Biol.*, Tome 28.

⁴⁶ Child, 1916, *Science*, Vol. 43.

kinds of cytoplasm. The appearance of a definite polarity might lead in some way to diffusion processes and the circulation of secretions resulting in further specializations and localizations. One stage seems to initiate the next stage in the series of processes that accompany the visible changes in the growth and development of the egg, and the character of these processes is of course due to the specificity of the protoplasm.

That the cytoplasm may exert a controlling influence upon the chromatin has been demonstrated in several instances. For example, we know that the chromatin-diminution processes during the early cleavage of both *Ascaris* and *Miastor* are controlled by the cytoplasm and that in these animals the germ-cell nuclei retain the full amount of chromatin because of the germ-cell cytoplasm they chance to encounter.

Probably the peculiar distribution of the chromosomes at certain stages in the life histories of certain aphids, phyloxerans, and Hymenoptera is also controlled by the cytoplasm. In the aphid, *Aphis saliceti*, the first maturation division is visibly differential both as regards the chromosomes and the cytoplasm.⁴⁷ The mitochondria congregate at one end of the dividing spermatocyte; this process is accompanied by a greater accumulation of cytoplasm at this end so that cell division results in one large cell containing all of the mitochondria and about two thirds of the cytoplasm, and one functionless small cell. The large cell also receives three chromosomes; the small cell only two.

The peculiar maturation divisions in the males of the honeybee⁴⁸ and hornet,⁴⁹ during which one ultimate spermatogonium gives rise to only one spermatozoon instead of the usual four, may also be the result of cytoplasmic control. The cytoplasm may likewise be responsible for the passing of a sex chromosome into the polar body during the maturation of the egg of certain aphids at the end of the summer season.⁵⁰ Such eggs must be fertilized

⁴⁷ Baehr, v, 1909, *Arch. für Zellf.*, Bd. 3.

⁴⁸ Meves, 1907, *Arch. mikr. Anat.*, Bd. 70.

⁴⁹ Meves and Duesberg, 1908, *Arch. mikr. Anat.*, Bd. 71.

⁵⁰ Morgan, 1909, *Journ. Exp. Zool.*, Vol. 7.

before they will develop, and always produce males. Many other peculiarities in the behavior of chromosomes that have been reported from time to time may also be due to the influence of the environment (cytoplasm), and there seems to be no reason why factors carried by the chromosomes should not be affected by the cytoplasm as well as are entire chromosomes.

By the interaction of Mendelian factors with the cytoplasm during the germ-cell cycle, it is even possible to explain the fact that "crossing over" occurs in the females of *Drosophila*, but not in the males.⁵¹ In the latter, the spermatocytes do not pass through a pronounced growth period, and hence there is comparatively little nucleo-cytoplasmic interaction, and since the cytoplasm carried by the sperm may be considered negligible, the factors borne by its chromosomes are not interfered with. In the female, however, there is ample opportunity for such interaction during the growth period, and factors at this time may be influenced by the cytoplasm or may influence the cytoplasm in such a way as to cause an irregular distribution of chromosomal factors.

To the writer the following conclusions seem justified. The insect egg at the time of maturation is a mosaic of differentiated cytoplasmic areas predetermined to develop into definite parts of the embryo. This organization has resulted from the interaction of nucleus and cytoplasm during the germ-cell cycle. Such interaction is taking place at all times, but is visible only when such processes as the protrusion of chromidia or chromatin-diminution occur. The many cases of cytoplasmic control over chromatin behavior, and the apparent failure of the factors for the characters commonly used by geneticists to influence the egg organization, indicate the importance of more careful studies of the genesis of this organization. The importance of such studies is emphasized by the possibility that they may help toward the solution of the problem of the method of evolution.

⁵¹ Morgan, Sturtevant, Muller, and Bridges, 1915, "Mechanism of Mendelian Heredity."

NEW FACTS AND VIEWS CONCERNING THE OCCURRENCE OF A SEXUAL PROCESS IN THE MYXOSPORIDIAN LIFE CYCLE.¹

DR. RHODA ERDMANN

OSBORN ZOOLOGICAL LABORATORY, YALE UNIVERSITY AND ROCKEFELLER
INSTITUTE FOR MEDICAL RESEARCH, DEPARTMENT OF
ANIMAL PATHOLOGY

THE classic observations of Balbiani, Bütschli and Thélohan on the myxosporidian development do not include the occurrence of a sexual process which comprehends the forming of a syncaryon in the life history of this protozoan group. Doflein (1898 and 1901) suggested *two* places in the life cycle of the myxosporidian where a caryogamy might probably take place. In the next period of investigations on myxosporidia the occurrence of this sexual process was stated by various authors, but they differ widely in the conception of the place in the life cycle in which the copulation occurs. Mercier, Awerinzew,

¹ For a clear understanding of the question under discussion, it is necessary to have a uniform nomenclature and to discard all terms which are only of historical value. Noyaux du sporoplasma, noyaux du germe, noyaux sporoplasmiques, germnuclei, Amöboidkeimkerne were to be discarded and gametonuclei, noyaux des gamètes, Gametenkerne were to be used. Instead of sporoplasma, sporoplasm or amöboidkeim, only such expressions as gametes, gametes and Gameten are admissible. Identical and adequate terms are capsulogenous cell, cellule capsulogène, Polkapselzelle. Valve cells, cellules valvaires and Schalenzelle should be used for those cells which form the membrane of each single spore; cellule d'enveloppe, envelope cells, and Hüllzellen should be used for those cells which form the membrane of the pansporoblast. In cases where only the nuclei of these cells are present, the terms noyaux d'enveloppe, envelope nuclei, Hüllenzellenkerne should be substituted. If the cellular origin of the pansporoblast membrane is not ascertained, envelope, membrane d'enveloppe or Hülle may be used. The terms Restkern, residual nucleus are misleading. Somatic residual nuclei or somatische Restkerne should be used, if the definition on p. 679 holds true. Here a new name would be of great value to eliminate the wrong analogies created by Doflein (1898, p. 309). The term "reduction nuclei" is only justified if the numerical reduction of chromosomes has been ascertained.

Auerbach and Parisi try to show that a real syncaryon formation takes place at the *onset* of spore formation. Other authors, Keysselitz, Schroeder and Auerbach, believe that only a plasmogamy can be pointed out at the beginning of spore formation, and that the union of the nuclei is effected either in the fully developed spore or in the young animal leaving the spore. The difference between these last two conceptions is theoretically without significance because the main part of copulation—the union of the nuclei—takes place *at the onset of the new life cycle of the myxosporidian*. Therefore it was of the utmost importance for decisive proof of this fact to find the copulation of the two gametonuclei inside the fully developed spore or in the young myxosporidian. Schroeder, 1909, observed the copulation of the two gametonuclei in the spore; Auerbach, 1907 and 1910, found young animals of *Myxidium bergense* with one nucleus. I was able to demonstrate young *Chloromyxum leydigi* which were experimentally produced by placing the two-nucleated spores on gall plates (Erdmann, 1911). Here, after a treatment with intestinal secretions of the host the young animals leave the spore. They are at first binucleate, later uninucleate. In my recent work, finished in 1913, which did not appear until 1917 in consequence of the war, I figured these young animals after fixation and staining. Also, Davis, 1915, though with some reserve, presents young *Sphaerospora dimorpha* which have left the spore and show the fusion of their two nuclei. Later the separation of the syncaryon into its vegetative and generative components takes place. Georgevitch, 1914, presents the development of the young animal in *Henneguya gigantea* in—as it seems—changed peculiar conditions. The spore is still inside of the cyst of the big “tumor-forming” tissue-parasite. The binucleate form becomes uninuclear and then the usual vegetative multiplication of the nuclei begins, which leads up to a *renewed* spore formation *inside* of the tumor cyst. In *Chloromyxum leydigi*, a gall-bladder parasite, no such

complicated process takes place. As the young uninucleated forms develop we see an animal with three nuclei, all of the same size. Later multiplication of these vegetative nuclei and the formation of big syncytial masses occur. The plasmatic bodies of these vegetative animals contain two kinds of round corpuscles, "Reservekörper" and "Farbträger." In my publication in the *Archiv für Protistenkunde*, 1917, I give proof that the "Reservekörper" consists mostly of glycogen and I point out that the glycogenous contents are used up during spore formation. The vegetative animal can multiply either by division or by forming small vegetative gemmules (Erdmann, 1911). The fact that inside the animal vegetative propagative bodies can arise, was verified by Davis, 1915, pp. 354-355, in *Sphaerospora dimorpha*.

Before the onset of spore formation a differentiation in the syncytial masses of *Chloromyxum leydigi* begins. We can distinguish parts in which the nuclei multiply and other parts where only the vegetative nuclei are seen widely scattered in the protoplasm. I called the first-mentioned areas "islands" (Erdmann, 1911) because in the living animal they rise above the surface of the vegetative plasmatic body. They are distinguished by their pale color and in stained preparations by their large number of small nuclei. At first all the nuclei in these islands are of the same size. Two nuclei with small cytoplasmic bodies approach each other and each cell divides up into a small and a big cell. The two small cells draw out in length and surround the two big ones, in this manner separating them from the other cells in the island. This quadruple group, two big cells and two small ones, is the starting point for the formation of the whole spore. The two big cells are gametocytes. These two gametocytes divide and form two gametes and two other cells which after a further division give rise to four cells—these four cells are the four capsulogenous cells. The whole spore contains, therefore, eight cells—four capsulogenous cells, two gametes and two cells which form the spore membrane. (Fig. 1.)

I mentioned before that the glycogen which was found in the vegetative body is used up in spore formation. The membrane of the spore, the polar threads and the darkly staining structureless lumps, which have been seen by all authors inside the sporoblast, consist of glycogen and stain as well by chromatin as by specific glycogen

stains. These lumps have been considered as "reduction nuclei" by various authors. They have also been called "Restkerne" or "residual nuclei." It may be emphasized for later discussion that they are glycogenous and not chromatic in *Chloromyxum leydigi*.

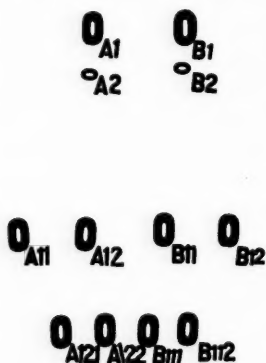


FIG. 1. *Chloromyxum leydigi*: A₂ and B₂ are the spore membrane forming cells; A₁₁ and B₁₂ are the gametes. A₁₂₁ A₁₂₂; B₁₁₁ B₁₁₂ are the capsulogenous cells.

Our knowledge of the sexual process in the myxosporidian life cycle since the investigations of Keysselitz, Schroeder, and others has been completed by recent authors: Georgevitch, 1914, Davis, 1915 and Mavor, 1916.

The best proof that *no* reduction takes place in the spore is given by Georgevitch (1914) and Davis (1915), who have been able to study the number of chromosomes in their specimens before and after spore formation. Both authors agree that the number of chromosomes is not changed before and after this phenomenon. Davis figures six chromosomes in *Sphaerospora dimorpha* and Georgevitch, investigating *Henneguya gigantea*, finds eight. These two facts: *first, that the number of chromosomes is not changed in spore formation* and, *second, that the so-called "reduction nuclei" inside the spore are really glycogenous bodies* which are used up in forming the membrane of the spore, polar threads, and spore membrane of *Chloromyxum leydigi*, positively prove that the real reduction must occur at a different place in the life cycle of *Sphaerospora*, *Chloro-*

myxum, and *Henmeguya*. The reduction consisting in the transformation of a diploid nucleus into a haploid or of a tetraploid one into a diploid can only occur at the beginning of the new life cycle *after* or *before* the union of the gametonuclei.

The further facts which Davis, 1915, presents in *Sphaerospora dimorpha* also tend to show that reduction could only take place at the above outlined place. The gametonuclei of this myxosporidian fuse together after it leaves the spore and form by one subsequent division *two* nuclei. One of these nuclei gives rise to *all* the cells of the later sporogenous body. The other of these two nuclei, distinguished by its size and structure, is the vegetative nucleus of the animal, the somatic "Restkern." *No other nuclei should be called "Restkerne" except when they represent the nucleus or nuclei of the vegetative myxosporidian body, which does not play a part in spore formation.* Such "Restkern" or "Rest-

TABLE I

Author	Species	Occurrence of Somatic Residual Nuclei in Sensu stricto. See Definition, p. 679	Occurrence of Hüllzellen Envelope Cells, Pan-sporoblast-membrane Forming Cells or Only Their Nuclei	Number of Valve Cells. They Are Division Products of Gametocytes
1. Awerinzew.	<i>Myxidium</i> sp.	One	—	2 for each spore
2. Auerbach...	<i>Myxidium bergense</i>	None (p. 26)	—	"
3. Davis.....	<i>Sphaerospora dimorpha</i>	One, seldom two	—	"
4. Awerinzew.	<i>Ceratomyxa drepanop-settae</i>	Two	—	"
5. Mavor.....	<i>Ceratomyxa acadensis</i>	Two	—	"
6. Erdmann...	<i>Chloromyxum lydigi</i>	Many	—	"
7. Davis.....	<i>Sphaerospora dimorpha</i> , polysporous form	Many	—	"
8. Auerbach...	<i>Myxidium bergense</i> , polysporous form	None	—	"
9. Parisi.....	<i>Sphaerospora cavdata</i>	(It may be that Auerbach has observed in his mono-, di- and polysporous forms only the propagative parts of the complete animal.) No facts mentioned	Two	"

kerne," the origin and fate of which agree with this definition, have been described by Awerinzew in *Ceratomyxa drepanopsetta*, and in *Myxidium* sp., by Davis in *Sphaerospora caudata*, and by Mavor in *Ceratomyxa acadiensis*.

In the disporous form, *Sphaerospora dimorpha*, two spores are found in the whole animal and the sporogenous body finally contains twelve cells—half of this number forms one spore (Fig. 2). These twelve cells are all

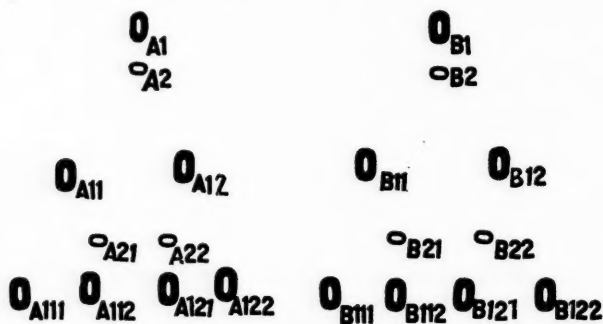


FIG. 2. *Sphaerospora dimorpha* (disporous form): A_{21} A_{22} and B_{21} B_{22} are the membrane-forming cells for each spore. A_{111} A_{112} ; B_{111} B_{112} are the two gametes in each spore. A_{112} A_{121} ; B_{112} B_{121} are the two capsulogenous cells in each spore.

division products of the *one* nucleus, the *sister* nucleus of which is the somatic "Restkern." The cells which form the spore membrane of each spore, lying independently in the vegetative body, arise by a very *late* division. Chromatic lumps which could be considered as "reduction nuclei" are lacking. It is easy to imagine why they are absent. *Sphaerospora dimorpha* lives in the urinary bladder and has therefore a metabolism which may not afford opportunity for an abundant glycogen formation. Also, as mentioned before, no reduction of the chromosomes takes place. These facts could be easily ascertained in consequence of the large size of the cells and chromosomes. Awerinzew's description of spore formation in *Ceratomyxa drepanopsetta* and Mavor's of *Ceratomyxa acadiensis* have many features in common, but Awerin-

zew's presentation differs from that of the other author in one important point. The cells *A* and *B* in Fig. 2 are said to be in *Ceratomyxa drepanopsettae* the products of a fusion of two cells. The reader has but to change the lettering of *A* in *AC* and of *B* in *BC* to see that the two copulae undergo changes identical with the two single cells in *Sphaerospora dimorpha*. Except this one point of difference—the beginning of spore formation—we note that the late division of the membrane-forming cell, the identical number of cells in each spore and the independence of each spore in the myxosporidian body characterize both species of *Ceratomyxa* (Awerinzew and Mavor) and *Sphaerospora* (Davis). It appears highly improbable that in two different species of *Ceratomyxidae* the basis of spore formation should be a copula (Awerinzew) or an univalent nucleus (Mavor).

Summarizing the known description of spore formation of those myxosporidia, in which *each spore is formed independently of the other in the somatic body*, and where no pansporoblast exists, we can demonstrate the following uniform features in all investigated forms.

1. Six cells or nuclei are used for the formation of each spore, when two polar capsules are present; eight, when four polar capsules are present.

2. The cells which form the spore membrane have a similar origin and are distinguished by the independence in which these cells develop as compared with the other constituents of the spore. Their mother cell is lying in a resting stage till the division of the gametocyte is finished, as described by Davis for *Sphaerospora dimorpha*, and by Awerinzew for *Ceratomyxa drepanopsettae*.

In *Myxidium* sp., where, according to the investigations of Awerinzew, either one, two or three spores are lying independently in the myxosporidian body, there is a very late division of the cell, the divisional products of which form the spore membrane as recorded by this author (Fig. 3). Here the one gametocyte divides into two cells, one of which by a late division gives rise to the two-

spore membrane-forming cells, the other forms the two capsulogenous cells and two gametes. We intentionally

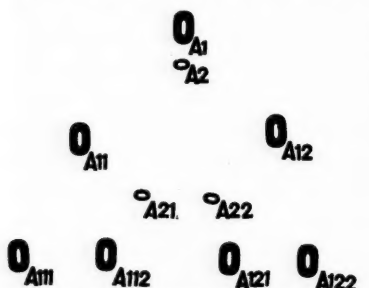


FIG. 3. *Myxidium* sp.: A_{21} and A_{22} are the spore membrane-forming cells (valve cells). A_{111} A_{112} are the gametes. A_{112} and A_{121} are the capsulogenous cells.

these chromatin lumps only as formations probably of glycogenous nature and as being used during membrane formation.

In Fig. 4 Auerbach's conception (Type I) of how the spore formation is effected in *Myxidium bergense* is represented. Auerbach believes, as stated before, that either a plasmogamy (Type I) or a real copulation (Type II) may be at the basis of spore formation. We will not discuss, for the present, how the bigger and smaller cells which are seen at the beginning of spore formation, arise. The latter divides once and the two divisional products form the spore-membrane-forming cells. The other cell divides twice to give rise to two gametes and two capsulogenous cells. The author does not

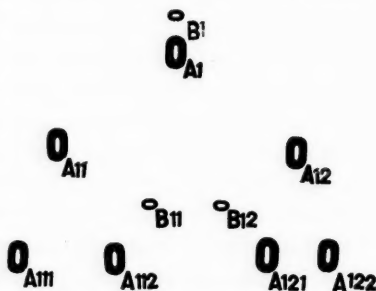


FIG. 4. *Myxidium bergense* (Auerbach Type I); B_{11} and B_{12} are the spore membrane-forming cells (valve cells). A_{111} and A_{112} are the two gametes. A_{112} and A_{121} are the capsulogenous cells.

especially mention the order in which these cells divide,

but nothing in his report contradicts the supposition that in *Myxidium* sp. and in *Myxidium bergense* there is a close analogy with Davis's and Awerinzew's observations. He also observes the elimination of chromatin and "eventuell Bildung von Restkernen" (p. 24). These are not incorporated in Fig. 4 for the same reasons I pointed out for *Myxidium* sp.

My observations in *Chloromyxum leydigi* show, furthermore, that the cells which form the valves of the spore do not play a part in the development of the final contents of the spore, but are here in this form (comp. Fig. 1) the products of the first division of each gametocyte. In the case of *Chloromyxum leydigi* two gametocytes form by one division the two spore-membrane-forming cells; in the three other species, *Myxidium* sp. (probably *Myxidium bergense*), *Ceratomyxa drepanopsettae* and *Sphaerospora dimorpha*, one gametocyte forms the one cell, the division products of which are transformed into the valves of the spore. But in all four species these cells have the sole purpose of forming the spore membrane.

After surveying the Figs. 1, 2 and 3, and having compared them, I have no doubt that the origin of the spore-membrane-forming cells is identical in the so-called monosporous and disporous forms. Advancing one step farther and taking into consideration those forms in which two gametocytes form the cells inside each spore (Fig. 1) we notice that in dealing with the origin and the position in the development of the spore, we have to add nothing. The spore-membrane-forming cells are distinguished by their early segregation from the gametocytes and their non-entering into the series of those cells which are included in the spore. The only difference is that these cells do not divide further; if they did, we could easily construct the disporous type of *Sphaerospora dimorpha* (Fig. 2). The same holds true for those species which form two spores in one pansporoblast (Fig. 4) and where two gametocytes are observed at the basis of spore formation (Keysselitz, Schroeder).

If we conceive these two cells in question (A_2 and B_2) which form in *Myxobolus pfeifferi* (Keysselitz) the pansporoblast membrane to divide once more and the last division inside the spore to be suppressed, we could have the type of *Sphaerospora dimorpha*.

According to Keysselitz, in *Myxobolus pfeifferi* each of the two gametocytes together with the small cell ap-

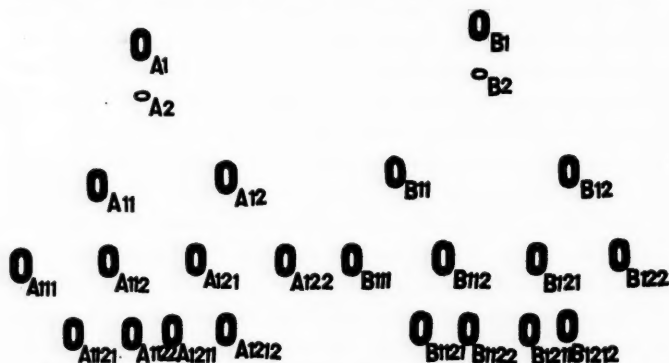


FIG. 5. General plan for polysporous disporoblastic forms. (Type Keysselitz.) A_2 and B_2 are the envelope-forming cells (pansporoblast-forming cells). A_{111} and A_{112} ; B_{111} and B_{112} are the two gametes in each spore. The products of the fourth division form valve cells A_{1121} , A_{1211} , B_{1121} , B_{1211} , and capsulogenous cells A_{1122} , A_{1212} , B_{1122} , B_{1212} .

proach each other and each divides up until six cells have arisen. Thus we have in all cases 14 cells, two of which have a different divisional capacity, for they stop dividing and the big cells form all the other cells which at the end compose the two spores. Their later fate is indicated in Fig. 5 and, though this does not concern us in this discussion, we should like to emphasize the fact that two gametes are always present in a certain stage of spore development. We are convinced that the cells A_2 and B_2 represent genetically in the pansporoblastic forms that cell formation which in all monosporous, disporous and polysporous species gives rise to the spore membrane itself (Figs. 1 to 4). These cells or their nuclei were observed by Keysselitz, Parisi, Lo Giudice, Auerbach (*Henneguya psorospermica*), Georgevitch, and with cer-

tain restrictions by Mercier and Schroeder. This cell couple (A_2 and B_2) should be called envelope cells or envelope cells nuclei when they do not fulfill their destin (Schroeder, probably Mercier). Even in those cases in which a pansporoblast membrane had not been discovered it might have been either overlooked or have been in evidence at the beginning of pansporoblast formation before the valves of each single spore had been developed. Later these take up the function of the Hüllzellen which make their retrogressive development plausible. These Hüllzellenkerne are neither Restkerne, nor reduction nuclei, nor somative residual nuclei. The term for residual nuclei of somatic nature (see definition, p. 679) has already been disposed of in *monosporous* and *disporous* forms and must be used in the same way in polysporous forms. In all forms which are polysporous and have many singly developing spores, the whole vegetative body which is not used up in spore formation has somatic "restkerne" or residual nuclei. As I pointed out in *Chloromyxum leydigi* the vegetative animal may die after spore formation together with the "restkerne." In this form the vegetative animal may prolong its life by forming internal buds, if it has reached a considerable size before and during spore formation. In all polysporous forms with pansporoblast, *i. e.*, the disporoblastic forms, we have to be very careful when applying the name of somatic residual nuclei. In those species which are not tissue parasites and sometimes have cystlike formations which are surrounded by gelatinous envelopes, we may find somatic residual nuclei, because it seems improbable that the whole vegetative body is used up for spore formation. I believe this to be the case in *Sphaeromyxum sabrazesi* and *Sphaerospora caudata*. Where no residual vegetative nuclei were observed, the investigators may not have studied the whole animal, but only the propagative parts of it which have left the vegetative body (Parisi, Fig. 3). Keeping this point in mind, later investigators may discover somatic residual nuclei in de-

generating stages analogous to those found in the monosporous, disporous and the non-disporoblastic polysporous forms, among the debris of the dying animals inside the gall and urinary bladder, as I have shown in *Chloromyxum leydigi*. In the so-called "tumor-forming" disporoblastic polysporous forms, no facts are known which show that somatic residual nuclei have been observed. The beginnings of cyst formation, however, have never been studied, and it is only at this stage that somatic residual nuclei may be seen, and not after the cyst is crowded with sporoblasts and spores. But even in the fully developed cysts there may be degenerating somatic residual nuclei which have escaped observation. The facts which Weissenberg found in *Glugea anomala* and *hertwigi*—two Microsporidia—seem to support my suggestion. But the Hüllzellen or Hüllzellenkerne of the myxosporidia are never identical with somatic nuclei. Their undisputed place in the development of the myxosporidia will soon be clear.

Before we proceed further in discussing the formation of the sporoblast membrane in those myxosporidian species in which the spores are formed in *pairs* inside one sporoblast, it may be recalled that several facts have been ascertained concerning the fully discussed spore-membrane formation in monosporous or disporous myxosporidian species: (1) The copulation of two gametes occurs during or after the two myxosporidia leave the spore. (2) No reduction takes place from the beginning of spore formation until the end, because the number of chromosomes remains the same (Davis and Georgevitch). (3) The darkly staining masses of "restkerne," "residual nuclei," "reduction nuclei" have been shown to be glycogenous and to be necessary for spore-membrane formation. (4) Membrane-forming cells or nuclei are set apart by different division intervals from the other cells of the sporoblast.

Now from the above given summary of the latest facts in monosporous or disporous forms, it is clear that they

are strictly *opposed* to all the views which maintain that a copulation or so-called syncaryon formation precedes spore formation. But they are all in accord with the investigations of all authors who have shown that there is no syncaryon formation, but merely a plasmogamy of two cells, without any copulation, at the onset of spore formation.

When I wrote the second part of my investigations on *Chloromyxum leydigi*, 1913, I pointed out that the facts which were presented by Auerbach, Mercier and Parisi, as proofs of the occurrence of a syncaryon formation just before the onset of spore formation, are not quite convincing. Their figures can easily be arranged in such a manner that the supposed syncaryon formation represents the division of gametocytes into a smaller cell, which in most all other known cases forms the membrane of the pansporoblast. (Compare Erdmann, 1917.) It is not necessary to repeat here the attempted revision and rearrangement of the figures of these authors. This same holds true for the syncaryon formation in *Myxidium bergense*, Type II (Auerbach) and *Ceratomyxa drepanopsettae* (Awerinzew). We will take it for granted that our views are correct as long as no new *facts* ascertained on smears—not sections—compel us to change our opinion.

As mentioned above, all authors who have shown that no syncaryon formation occurs, but that a plasmogamy of two cells without any nuclear fusion occurs at the onset of spore formation, can agree with us that the sexual process is going on at the beginning of the new life cycle. Auerbach and Parisi do not convince us that the figures which represent the so-called caryogamy can not be considered as the dividing of the gametocyte in the two cells. In accordance with the facts and interpretations, Keyselitz, Schroeder, Lo Giudice, Erdmann, and Georgevitch uphold the view that *no* merging of two cells or two cell pairs takes place to form the couples of cells which are later considered as a quadruple group of the growing spore. By comparing the series of figures of all those drawings

which are supposed to prove the merging of two cells, they can, as said before, be interpreted as the division of one cell into two. The larger of these cells, wrongly called macrogametocyte according to the cell fusion theory, has divided and formed the cell wrongly called microgametocyte.

This "microgametocyte" and its division products after one division, or these "microgametocytes" in cases where two gametocytes are observed at the onset of spore formation (Keysselitz, Schroeder, and Erdmann) now form, according to all known investigations, the pansporoblast membrane. Just as we could point out in disporous forms the uniformity of the origin of the spore-membrane forming cells (Figs. 2, 3 and 4) so we can do the same for the pansporoblast membrane and its nuclei in the following forms: *Myxobolus pfeifferi* (Keysselitz), *Myxobolus ellipsoides* (Lo Giudice), *Sphaeromyxa sabrazesi* (Schroeder), *Sphaerospora caudata* (Parisi), *Henneguya gigantea* (Georgevitch), and *Henneguya psorospermica* (Auerbach); all following Fig. 5, provided we do not take into consideration the origin of the cells *A*, *A*₂ and *B* and *B*₂. Keysselitz and Schroeder's views, except one contradictory point, are exactly represented by Fig. 5, but there are differences mentioned by the other authors. Still we make the generalization that there is one and the same plan of spore formation in the pansporoblastic myxosporidia though we know that facts are reported which do not fit in with our view. *We hold the opinion that it is permissible to rearrange the observed facts, because all interpretations have been gained by piecing together and arranging facts according to the theoretical viewpoint of the authors.* No continued observation of spore formation in the living animal has been possible. Also we are allowed to add facts ascertained in other species if the authors have only considered sections and not smears. Sections are misleading because the whole quadruple group can not always be seen on the same section and the origin of the small cell from

the big cell can not be traced without doubt. Therefore, most investigators have lately used smears to get a fuller and more correct view of the origin of the different cells from each other. It is astonishing how scanty the details appear when one considers the formation of the quadruple group in Auerbach's, Lo Giudice's, Parisi's and Georgevitch's presentations. Mercier's Figs. 19-27, Plate 1; Auerbach's Figs. 8a-15, Plate 2; Lo Giudice's Figs. 29-34, Plate 1; Parisi's Figs. 13-18, Plate 16; and Georgevitch's Figs. 32-35, Plate 1, do not show each single step of this important process. Connecting stages are missing. Therefore, one is allowed to interpret differently *their* presented facts, as I have done in Fig. 5. In Table II

TABLE II

I		II	
<i>Myxobolus pfeifferi</i> Mercier.		<i>Sphaeromyxum sabrazesi</i> Schroeder.	
Cell A (copula) forms all other 12 cells of the pansporoblast and the two "Hüllzellenkerne."		A_3	B_2
		A_1	B_1
		All cells divide up to form the 12 pansporoblastic cells and the two "Hüllzellenkerne."	
<i>Myxobolus ellipsoides</i>	<i>Henneguya gigantea</i>	<i>Henneguya psorospermica</i>	
Lo Giudice.	Georgevitch.	Auerbach (Type I).	
	A_2	A_{22}	
	A_1	A_{12}	
		Cells A_2 and A_{22} do not form any of the other 12 cells of the pansporoblast.	

we can study the different opinions from the authors' point of view. Lo Giudice, Georgevitch, Auerbach, Keyselitz, and Schroeder are alike in interpreting that the two cells which develop into the pansporoblastic membrane or nuclei are separated very early from the other cells. They never intermingle with those cells inside the spore-membrane (except according to Schroeder). They can not, therefore, be microgametocytes and in consequence they have nothing whatsoever to do with a sexual phenomenon. This adds strong support to our view that

the sexual process is at the beginning of the new life cycle.

I do not wish to veil the great discrepancy between the conception of Mercier (Fig. 6) and the other authors

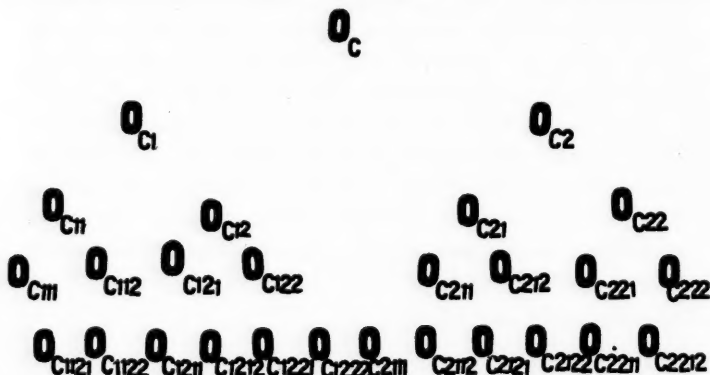


FIG. 6. *Myxobolus pfeifferi*, Mercier: C_{111} and C_{222} are the celles d'enveloppe of this author, which represent the false "Restkerne" or "reduction nuclei" of other authors. The products of the fourth division form, for each spore, the gametes, the valve cells and the capsulogenous cells.

mentioned. All cells are products of a copula, and there is no setting apart of the pansporoblast-membrane-forming cells or nuclei, though later they appear at the accustomed places between the two spores (Plate I, Figs. 31, 32). I shall not risk an interpretation, but think a new investigation on this same subject might be very promising and result in the desired uniformity. I think it highly probable that in all pansporoblastic forms the spore development follows the Keysselitz-Schroeder interpretation: that two gametocytes form the basis of the spore formation. But even if one believes that the quadruple group is not formed by two but by one cell pair, the principal point is not changed. It is indifferent for the theoretical interpretation whether a segregation of the second cell pair from the first takes place, and both then form the quadruple group, or whether two cell pairs approach each other and form the quadruple group. The paedogamy is merely a closer one in the first case.

To summarize: In the observed myxosporidian species

with pansporoblast (exception, Mercier, *Myxobolus pfeifferi*), the first division products of the gametocyte or gametocytes form the pansporoblastic membrane or, if degenerated, its nuclei. This division is a heteropole division and forms highly chromatic small cells or nuclei which never intermingle with the cells inside the pansporoblastic membrane (exception, Schroeder).

On the basis of these facts, we need only state that the heteropole division has no connection whatsoever with a reduction division, as Keysseltz tentatively suggested. This conspicuous division produces the Hüllzellen or Hüllzellenkerne, and it may not be impossible that in the case of *Sphaeromyxa sabrazesi* these small chromatin cells do not intermingle with the others and divide up, though the author mentions it on page 366. They originate, according to Schroeder's second interpretation, in the same manner as most authors describe, but divide together with the big cells until twelve cells are present in the pansporoblast; they then take their accustomed place inside the sporoblast membrane and are easily recognizable. This apparent exception merits further investigation.

We maintain our conclusion that the Hüllzellenkerne or the Hüllzellen are identical with the spore membrane forming cells of the non-disporoblastic polysporous forms. They have the following features in common: they are the first division products of the gametocytes; they do not intermingle with the other cells inside the spore; they form the envelope, in one case of the spore, in the other of the pansporoblast. They are neither somatic residual nuclei nor Restkerne nor reduction nuclei. They are cells which have a tendency to degenerate in some disporoblastic forms when their functions are taken up at an early period by the valve cells or spore-membrane-forming cells.

It remains, as the last part of our discussion, to deal fully with the significance of those darkly staining masses which have been described as "Reductionskerne" or "Restkerne" "inside the sporoblast." In the following table we give a short survey of the known facts.

A survey of Table III brings out clearly certain facts. When darkly staining masses are observed inside the

TABLE III

Author	Species	Occurrence of Chromatic Bodies Inside the Spore Interpreted as Reduction
1. Awerinzew	... <i>Myxidium</i> spec.	Seldom distinct small nuclei, generally "Zwei ziemlich grosse Chromatinkügelchen" (p. 201).
2. Auerbach <i>Myxidium bergense</i>	Diffusion of chromatin or formation of two "restkernartigen Gebilden" (p. 20).
3. Davis <i>Sphaerospora dimorpha</i>	Formation of round chromatic bodies.
4. Awerinzew	... <i>Ceratomyxa drepanosetæ</i> ..	Infiltration of chromatic small bodies into the cytoplasm before the spore membrane includes the gametocytes after the supposed copulation. Note here the later formation of spore membrane.
5. Mavor <i>Ceratomyxa acadiensis</i>	Formation of round chromatic bodies at the onset of spore formation (Fig. 10) which are later resolved.
6. Erdmann <i>Chloromyxum leydigi</i>	Formation of several large, deeply staining bodies which disappear after the spore membrane is formed.
7. Parisi <i>Sphaerospora caudata</i>	Formation of small, deeply staining bodies before the spore membrane includes the gametocytes after the supposed copulation (Fig. 15).
8. Davis <i>Sphaerospora dimorpha</i> , polysporous form	No diffused infiltration of chromatin observed, also no formation of round chromatic bodies.
9. Auerbach <i>Myxidium bergense</i> , polysporous form	Formation of two "restkernartigen Gebilden" or diffusion of chromatin.

10. Keysselitz ... *Myxobolus pfeifferi* One to four round chromatic, deeply staining bodies, disappearing after the valves of the spores are formed (p. 261).
11. Mercier *Myxobolus pfeifferi* Diffusion of small chromatic bodies into the cytoplasm (Figs. 33, 34) after the sporoblast membrane is formed, also the same fact stated after the supposed copulation (Figs. 22, 23).
12. Lo Guidice ... *Myxobolus ellipsoides* Several round chromatic, deeply staining bodies, which are not observed after the spore membrane is formed (Figs. 41, 42).
13. Schroeder *Sphaeromyxum sabrazesi* ... Chromatic, deeply staining bodies which are not observable when spore membrane is formed. (Comp. Figs. 30, 32 with Figs. 33, 34.)
14. Auerbach *Henneguya psorospermica* .. To judge after Figs. 6 to 18, extrusion of a chromatic body in cytoplasm. (Note, only sections to judge from.)
15. Georgevitch .. *Henneguya gigantea* Four deeply staining chromatic bodies called by the author degenerated nuclei.

spore, they disappear after the spore membrane is formed. It is proved that in *Chloromyxum leydigi* they are of glycogenous nature as well as the spore membrane itself and the polar threads. In some cases their number is irregular. These chromatic lumps may be products of nuclear division, but the true chromosomes have not been found. Those authors (Mercier, Awerinzew, Parisi) who believe they have shown a syncaryon forming, have also observed an extrusion of chromatin immediately after the union of the supposed micro- and macro-gametocyte. In Mercier's case a second diffusion of round bodies is shown *inside* the spore which corresponds with the facts observed in other species. *Ceratomyxa drepanopsetta*

(Awerinzew) has only an extrusion of chromatin before the syncaryon formation, while in *Myxobolus ellipsoides* (Parisi) it occurs immediately after this phenomenon. These exceptions in the series, *i. e.*, that inside the spore no chromatin diffusion is observed, may be due in the case of Parisi to a limited number of studied forms and in the case of Awerinzew to the fact that the spore membrane in *Ceratomyxa* is formed very late. Yet these exceptions do not prevent the final statement that the darkly staining chromatic masses in the spore are not reduction nuclei, or restkernartige Gebilde, but play an important part in the development of the spore membrane.

The whole trend of our critical review leads up to the following conclusions:

1. Reduction in myxosporidia has thus far not been discovered.
2. The so-called reduction nuclei inside the spore are chromatic or glycogenous masses, which serve the spore-membrane formation.
3. The so-called residual nuclei of the disporoblastic forms can not be considered as identical with the somatic residual nuclei of the mono-, di- or poly-sporous non-disporoblastic forms. They are the functionless nuclei of the envelope cells of the disporoblastic forms.
4. The envelope cells can by their origin only be compared with those cells in the mono-, di- or polysporous nondisporoblastic forms which later give rise to the valve cells.
5. The somatic residual nuclei are well-defined in mono-, di- or poly-sporous nondisporoblastic myxosporidia. Their analogy has not thus far been discovered in disporoblastic polysporous forms.

LITERATURE

(For literature before 1910 compare Auerbach, M., "Die Cnidosporidien," Leipzig, 1910)

Auerbach, M. 1912. Studien über die Myxosporidien der norwegischen Seefische und ihre Verbreitung. *Zool. Jahrb. Abt. f. Syst.*, Vol. 34, pp. 1-50.

- Davis, H. S. 1916. The Structure and Development of a Myxosporidian Parasite of the Squeteague, *Cynoscion regalis*. *Journal of Morphology*, Vol. 27, pp. 333-346.
- Erdmann, Rh. 1911. Zur Lebensgeschichte von *Chloromyxum leydigi*, einer miktosporen Myxosporidie (Teil I). *Arch. f. Protistenkunde*, Vol. 24, pp. 149-162.
- Erdmann, Rh. 1917. *Chloromyxum leydigi* und seine Beziehungen zur anderen Myxosporidien (Teil II). *Arch. f. Protistenkunde*.*
- Georgevitch, I. 1908. Sur le cycle évolutif chez les Myxosporidies. *C. R. A. Sc. P.*, T. 158, p. 190.
- Georgevitch, I. 1915. Etude du cycle évolutif chez les Myxosporidies. *Arch. d. Zool. exp. et gen.*, T. 54, pp. 388-409.
- Lo Giudice, P. 1911. Sullo sviluppo del *Myxobolus ellipsoides* Thel. *Riv. mens. Pesca Idrobiologia*, Anno 6 (13).
- Lo Giudice, P. 1912. Studi sui Cnidosporidi. Pavia, pp. 1-88.
- Mavor, I. W. 1916. On the Life History of *Ceratomyxa acadensis*, a New Species of Myxosporidia from the Eastern Coast of Canada. *Contrib. Zool. Lab. of the Museum of Compar. Zoology at Harvard College*, No. 269, pp. 551-578.
- Nemeczek, A. 1911. Beiträge zur Kenntnis der Myxo und Microsporidien der Fische. *Archiv. f. Protistenkunde*, Bd. 22, pp. 143-169.
- Parisi, B. 1910. *Sphaerospora caudata* n. sp. *Zool. Anzeiger*, Bd. 36, pp. 253-254.
- Parisi, B. 1913. Sulla sphaerospora caudata. *Atti della Società Italiana di Scienze Naturali*, Vol. 51, pp. 1-11.

* In consequence of the war I did not see my own reprint printed in the Archiv für Protistenkunde so it is impossible to add volume and page reference.

EVIDENCE FOR THE DEATH IN UTERO OF THE HOMOZYGOUS YELLOW MOUSE¹

HEMAN L. IBSEN AND EMIL STEIGLEDER

CUENOT (1905) and Castle and Little (1910) have presented conclusive evidence that yellow mice are always heterozygous and hence cannot be made to breed true. Their combined results show that when yellows are mated together the proportion of yellows to non-yellows in the offspring is almost exactly 2:1 instead of the usual 3:1 ratio resulting from the mating of heterozygotes. Castle and Little seem justified on this account in assuming that the homozygous yellows are not viable, especially since the size of litter from the yellow \times yellow mating is markedly smaller than that obtained from yellow \times non-yellow or non-yellow \times non-yellow matings.

Until quite recently no attempt had been made to determine embryologically the actual fate of the homozygous yellows. Since the present investigation was begun, however, Kirkham (1917) has published a preliminary statement of the results of such a study. His results, presented only in abstract, show that of 69 embryos from yellow \times yellow parents, 26 or 37.8 per cent. were degenerating. For "non-yellows" he used albinos.² Of

¹ Papers from the Department of Experimental Breeding, Wisconsin Agricultural Experiment Station, No. 11. Published with the approval of the Director of the Station.

[The problem of the fate of the homozygous yellow mouse was undertaken at my suggestion during the summer of 1916 by Mr. Steigleder and the experimental work on which the present paper is based was done entirely by him. As he was, however, unable to complete the problem the accumulated material and records were turned over to Dr. Ibsen, who has checked all records with the preserved embryos and is alone responsible for the tabulation, interpretation and presentation of the results.—L. J. Cole.]

² This selection of albinos for "non-yellows" was unfortunate, since they apparently were not tested genetically, and hence may or may not have carried the factor for yellow. From the fact that the proportion of dead embryos was markedly different we may accept Kirkham's assump-

the 84 embryos from albino parents, only 2, or 2.3 per cent., were degenerating. This makes it seem quite probable that the homozygous yellow zygote develops for a time and then dies.

In our study no attempt has been made to investigate the very early stages, as was done by Kirkham, but a large number of embryos have been obtained from non-suckling females pregnant from 13 to 19 days.³ In all 688 embryos have been examined. These have been obtained from (1) yellow females mated to yellow males, (2) yellow females mated to non-yellow males (chocolates), (3) non-yellow females (chocolates) mated to yellow males, and (4) non-yellow females mated to non-yellow males. In this last mating most of the parents were self blacks.

During the investigation two distinct types of dead embryos were encountered, (1) those in which development had ceased shortly after implantation, corresponding to those described by Kirkham, and (2) a few which seem to have developed normally till about the thirteenth day and then died, presumably because of overcrowding in the uterus. These latter were characterized by their dead, yellow appearance and smaller size as contrasted with the living pink color and larger size of the normal embryos. The first kind has been designated "dead embryos A" in the tables, while the second kind is classed as "dead embryos B." We are primarily concerned with "dead embryos A," and it is to be understood that reference is to this type unless specifically stated otherwise.⁴ Similarly, by "living embryos" we mean those which

tion that they were really genetically non-yellows, though this brings the argument dangerously close to reasoning in a circle. In our work a number of albinos were mated both to yellows and to non-yellows, but since the genetic constitution of the albinos was not sufficiently established in most cases, the embryos from these matings are not included in our tabulations.

³ The normal duration of gestation in the mouse is about 21 days, but it is often less.

⁴ We are indebted to Dr. Alva Wilson for sectioning for us dead embryos from each of the four types of matings. Microscopical examination of these has verified our previous conclusions as to their character.

were obviously alive when the mother was killed. As the females were killed by chloroforming, all embryos were usually dead by the time they were examined. The following tables give the results obtained. Each of the first four tables represents one of the four types of matings described above. In a few instances there has been some uncertainty as to the exact stage of gestation at which the embryos were removed, but in such cases this has been determined approximately by means of weights of the living embryos.

TABLE I
YELLOW ♀ × YELLOW ♂

Stage of Gestation	No. of Litters	Living Embryos	Dead Embryos A	Dead Embryos B	Percentage Dead Embryos A	Average Size of Litter	
						Living Embryos	Total Embryos
13 days.	5	33	14		29.8	6.6	9.4
14 days.	6	50	9		15.3	8.3	9.8
15 days.	4	26	10	1	27.0	6.5	9.3
16 days.	7	42	13	2	22.8	6.0	8.1
17 days.	5	25	4		13.8	5.0	5.8
18 days.	5	27	13		32.5	5.4	8.0
19 days.	1		4		100.0		4.0
Total.	33	203	67	3	24.54	6.15	8.27

TABLE II
YELLOW ♀ × NON-YELLOW ♂

Stage of Gestation	No. of Litters	Living Embryos	Dead Embryos A	Dead Embryos B	Percentage Dead Embryos A	Average Size of Litter	
						Living Embryos	Total Embryos
13 days.	5	34	12		26.1	6.8	9.2
14 days.	6	56	1		1.8	9.3	9.5
15 days.	4	35	4		10.3	8.8	9.8
16 days.	2	14	8		36.4	7.0	11.0
17 days.	3	25	1		3.8	8.3	8.7
18 days.	3	17	1	1	5.3	5.7	6.3
19 days.	1	10				10.0	10.0
Total.	24	191	27	1	12.33	7.96	9.13

The data presented agree in the main with Kirkham's, but our percentage of dead embryos A, from the yellow × yellow mating (Table I) is considerably less than his, while our percentage from the non-yellow × non-yellow

mating (Table IV) is somewhat higher. Theoretically, if we assume that a certain proportion of embryos of other gametic composition die from unknown causes in mice of all colors and that all the homozygous yellows are

TABLE III
NON-YELLOW ♀ × YELLOW ♂

Stage of Gestation	No. of Litters	Living Embryos	Dead Embryos A	Dead Embryos B	Percentage Dead Embryos A	Average Size of Litter	
						Living Embryos	Total Embryos
13 days.....	1	2	1		33.3	2.0	3.0
14 days.....	1	6				6.0	6.0
15 days.....	2	15				7.5	7.5
16 days.....	2	20	1	1	4.5	10.0	11.0
17 days.....	2	11				5.5	5.5
18 days.....	1	8	1		11.0	8.0	9.0
19 days.....							
Total.....	9	62	3	1	4.55	6.89	7.33

TABLE IV
NON-YELLOW ♀ × NON-YELLOW ♂

Stage of Gestation	No. of Litters	Living Embryos	Dead Embryos A	Dead Embryos B	Percentage Dead Embryos A	Average Size of Litter	
						Living Embryos	Total Embryos
13 days.....	3	22	3		12.0	7.3	8.3
14 days.....	3	28	1		3.4	9.3	9.7
15 days.....							
16 days.....	3	22		2		7.3	8.0
17 days.....	3	19				6.3	6.3
18 days.....	3	26	3		10.3	8.7	9.7
19 days.....	1	4				4.0	4.0
Total.....	16	121	7	2	5.38	7.56	8.13

represented as dead embryos, then the difference between the percentages of dead embryos from the yellow × yellow mating and the non-yellow × non-yellow mating should be approximately 25 per cent. In our results the difference is 19.2 per cent., while in Kirkham's it is 35.5 per cent. Neither of these is especially close to the expected percentage. If, however, his results and ours are combined the difference is 23.0 per cent., which is close to expectation.

If instead of comparing yellow × yellow with non-yellow × non-yellow only, all matings other than yellow

× yellow are combined for this purpose, it is found that the dead embryos in these combined matings constitute 8.9 per cent. of all embryos present. Subtracting this from 24.5, the per cent. of dead embryos in the yellow × yellow mating, the difference is only 15.6 per cent. When all these results are combined with those of Kirkham the difference is still only 19.4 per cent., which is considerably lower than that obtained by using only the classes considered by him. An attempt will be made farther on to explain this deficiency.

Tables II and III, representing the reciprocal crosses of yellow × non-yellow, show marked contrasts in several respects. In the yellow ♀ × non-yellow ♂ mating (Table II) 12.3 per cent. of the embryos were dead, while in the non-yellow ♀ × yellow ♂ mating (Table III) the percentage is only 4.5 per cent. For both matings the percentage of dead embryos theoretically should be the same as in the non-yellow × non-yellow mating (Table IV), since in neither case is there the possibility of any of the offspring being homozygous yellows. The percentage in Table III is approximately the same as in Table IV, but in Table II it is much higher. It is well known that yellow females tend to take on more fat than females of other colors. There is a possibility that this physiological difference may also in some way influence the production of a greater number of dead embryos *A*. This is offered merely as a suggestion, since there is no direct evidence for or against it.

In discussing the stages of pregnancy in which the dead embryos are to be found, Kirkham makes a statement which does not accord with our observations. He says: "No degenerating embryos have been found in either white [albino] or yellow mice pregnant more than sixteen days." He believes complete resorption of the degenerating embryos has taken place by the end of the sixteenth day. Table V, which is a summary of all our four types of matings, indicates that in our material there is no marked decrease in the percentage of dead embryos toward the end of pregnancy.

TABLE V
RELATION OF DEAD EMBRYOS TO STAGE OF PREGNANCY FOR THE FOUR TYPES
OF MATINGS

Stage of Gestation	Dead Embryos <i>A</i>	Total Embryos (Including <i>B</i>)	Percentage of Dead Embryos <i>A</i>
13 days.....	30	121	24.8
14 days.....	11	151	7.3
15 days.....	14	91	15.4
16 days.....	22	125	17.6
17 days.....	5	85	5.9
18 days.....	18	97	18.6
19 days.....	4	18	22.2
Total	104	688	15.12

As previously stated, Cuénot, and Castle and Little noted that the average number per litter is less from yellow females mated to yellow males than from any other type of mating. Our results, summarized in Table VI, bear this out. Here it will be noted that the average number of living embryos is less for mating 1 than for any

TABLE VI
LITTER SIZE FOR THE DIFFERENT TYPES OF MATINGS

Type of Mating	No. of Litters	Average Number of Embryos per Litter		
		Living Em- bryos	Dead Embryos <i>A</i> and <i>B</i>	Total
1. Yellow ♀ × yellow ♂.....	33	6.15	2.12	8.27
2. Yellow ♀ × non-yellow ♂.....	24	7.96	1.17	9.13
3. Yellow ♂ × non-yellow ♀.....	9	6.89	0.44	7.33
(2 and 3 combined).....	33	7.67	0.97	8.64
4. Non-yellow ♀ × non-yellow ♂...	16	7.56	0.57	8.13
(2, 3 and 4 combined)	49	7.36	0.84	8.47

of the other types of matings singly or combined. It is evident that the average number of living embryos per litter represents those which would in the regular course of events have been born alive. In the yellow × yellow mating the average is only 6.15,⁵ while for the other matings combined it is 7.63. The average number when the dead embryos are also included should be approximately the same in both cases, and this proves to be true, being 8.27 for yellow × yellow and 8.47 for all other matings.

⁵ Two of the litters in this mating were made up entirely of dead embryos *A* and therefore had no living embryos. So far as living embryos are concerned their size would be 0, and they have been included as such with the other litters produced by this mating.

During 1916-17, the period at which the embryological study was carried on, numerous living litters of mice were born in the laboratory. Miss Sarah V. Jones, who was also working with mice, has generously furnished data from her own breeding experiments, and these have been incorporated with ours. Some of the mice in her experiments were black-and-tans, but as Dunn (1916) has shown that these are a form of yellow and are also always heterozygous, they have been classified as yellows. Table VII shows the average size of litters for the various types of matings.

TABLE VII
AVERAGE SIZE OF LITTERS BORN DURING 1916-17
(Figures in parentheses indicate the number of litters.)

Type of Mating	Average Number of Mice per Litter	
Type of Mating	At Birth	At Time Color was Recorded
1. Yellow ♀ × yellow ♂	5.36(140)	4.55(121)
2. Yellow ♀ × non-yellow ♂	6.21(88)	5.63(71)
3. Yellow ♂ × non-yellow ♀	7.02(50)	5.95(46)
(2 and 3 combined)	6.51(138)	5.76(117)
4. Non-yellow ♀ × non-yellow ♂	6.88(42)	5.49(37)
(2, 3 and 4 combined)	6.59(180)	5.69(154)

It will be seen that there is a deficiency of litter size from the yellow × yellow mating here as in the embryological material. Theoretically the average size of litter from yellow parents should be 75 per cent. of that from any of the other matings. Both Cuénot, and Castle and Little, however, have found it to be above 80 per cent. Our results are in accord with these findings.⁶ Table VIII shows the percentages found by the various investigators. In order to make them comparable with the others our results as given in the table are for the living litters only and do not include the data from the embryological investigations.

⁶ Miss Durham (1911) found very little difference in average litter size between the offspring from yellows × yellows and the offspring from matings where at least one of the parents was a non-yellow. She says: "Only mice which lived long enough to have their colors determined are included in these averages." It seemed possible to us that she had not found any

TABLE VIII

PROPORTIONATE SIZE OF LITTERS FROM YELLOW \times YELLOW AND YELLOW \times NON-YELLOW MATINGS

(Figures in parentheses represent number of litters.)

Authority	Average Size of Litter		Ratio of "Yellow \times Yellow" to "Yellow \times Non-yellow"
	Yellow \times Yellow	Yellow \times Non-yellow	
Cuénot (1909).....	3.38(50)	3.74(50)	90.37 %
Castle and Little (1910).....	4.71(277)	5.57(325)	84.63 %
Ibsen and Steigleder, 1917.....	5.36(140)	6.51(138)	82.33 %

The most striking fact brought out in this table is that as the average size of the litters increases the ratio tends to decrease and therefore to approach 75 per cent. It would seem from this that if one could secure a race of mice having a high enough average per litter the theoretical percentage could be obtained. Using Table VIII as a basis for computation, such a race should average approximately 10 young per litter in the non-yellow \times yellow mating and consequently about 7.5 per litter for the yellow \times yellow mating. It is not probable that a race of this sort exists.

Various theories have been advanced to explain this unexpectedly large litter size in the yellow \times yellow mating. Two suggested by Castle and Little will be considered here. Both take as their starting-point that "the perishing of a pure [homozygous] yellow zygote makes possible the development of a certain number of *other* fertilized eggs." The explanation follows: "Two ways may be suggested in which this might come about. First, more eggs may normally be liberated at an ovulation than there are young born subsequently. In that case, failure of some eggs to become attached to the uterus may make the chances greater that the remainder will become at-difference because the size of litter had not been recorded at birth. Our records have been gone over and the litter size found at the time the colors were recorded. Some litters did not live long enough to be recorded in this manner and hence could not be included. By referring to Table VII one can see that our results do not at all agree with Miss Durham's. As a matter of fact, in our material the percentage relation is lower than when the litter size was computed at birth, being almost exactly 79 per cent. when "yellow \times yellow" is compared with "non-yellow \times yellow."

tached, or the perishing of some may make the chances greater that the rest will successfully complete their development. Or secondly, the production of a relatively small number of young at one birth may lead indirectly to more free ovulation subsequently, and so to the production of a large litter at a second birth."

It will be seen in the first place that the above theories are based on false premises. It was not known at the time these theories were proposed that the homozygous yellow zygote does *not* perish in the sense of disintegrating and finally disappearing. It merely ceases to develop after a certain stage has been reached, and then remains more or less stationary till parturition. It might still be maintained that since these undeveloped zygotes take up very little room there would still be the possibility for the "other" zygotes to develop. In that case the average number of total embryos (including dead embryos *A* and *B*) should be greater in the yellow \times yellow mating than in the non-yellow \times yellow mating. Our data, presented in Table VI, indicate that this is not the fact. The other theory that "the production of a relatively small number of young at one birth may lead indirectly to more free ovulation subsequently, and so to the production of a larger litter at a second birth" still remains a possibility so far as our data are concerned.

There are several other possibilities which seem worthy of consideration. Instead of looking for causes that tend to *increase* the size of litters from yellows \times yellows it may be profitable to determine if possible causes that may *decrease* the size of litters from the non-yellow \times yellow mating. The first and most obvious possibility is that overcrowding in the uterus may have this effect by causing the death of some of the embryos. However, none of our results bear this out. In the first place there are proportionately just as many dead embryos *B* (whose death is probably caused by overcrowding) in the one type of mating as in the other, and in the second place this would mean that for our race of mice, having a high average litter size, there should be a proportionately

large death rate due to overcrowding, and this would tend to increase the percentage relation between "yellow \times yellow" and the "non-yellow \times yellow" instead of decreasing it, which actually is the case.

We know that dead embryos *B* do not materially decrease the litter size in the non-yellow \times yellow mating, but on the other hand we have clear evidence that dead embryos *A* have a decided effect in this respect. The only manner we could postulate in which this could affect the litter-size percentage relation for the two contrasted types of matings would be to assume that in the yellow \times yellow mating dead embryos *A* are due almost entirely to the fact of their being homozygous yellows, while in the non-yellow \times yellow mating separate agencies are at work producing an appreciable number of deaths. If a more careful examination of dead embryos *A* should reveal rather easily distinguishable differences this explanation could be tested.

There is still another explanation which so far as we know has never been suggested, and which has more evidence in its favor than any of the others proposed. When yellows are mated to yellows it is to be expected that some of the litters, especially if they are small, will consist entirely of homozygous yellows. Since these do not complete development, the entire litter will be composed of dead embryos *A* and consequently there will be no living embryos born at parturition. Such a litter will therefore be of 0 size. In our embryological investigation two litters consisting entirely of dead embryos *A* were found in the yellow \times yellow mating and none in the other types of matings. Even with these two litters of 0 size in the yellow \times yellow mating the average size of litter as compared to the litters of the non-yellow \times yellow mating was not sufficiently low to bring the percentage relation down to 75 per cent. It is, however, 80.18 per cent., which is appreciably lower than the percentage obtained from litters of animals allowed to give birth to their young. (See Table VIII.) In the latter case it would naturally be impossible to detect the litters of 0 size.

In our embryological study there were 33 litters in the yellow \times yellow mating. Of these, as previously stated, two consisted entirely of dead embryos *A*. This means that for 31 litters containing living embryos there were two that did not, or 6.45 per cent. If we assume a like

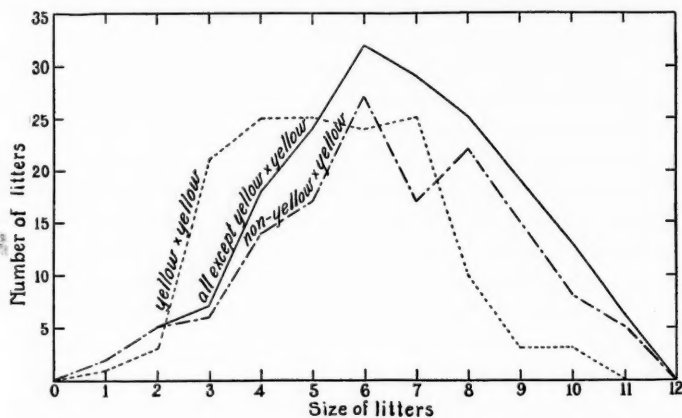


FIG. 1.

percentage of litters of 0 size with our 140 normally born litters from "yellow \times yellow" (Table VIII) the total number of litters would be increased to 149 and their average size would be 5.04 instead of 5.36. Taking 5.04 as a basis for comparison with 6.51, the average size of litters from the non-yellow \times yellow mating, we find that the percentage relation is 77.42 per cent. This is reasonably close to 75 per cent., the expected percentage. In this connection it may be well to call attention to a fact, previously stated, that as the average litter size increases the percentage relation tends to approach 75 per cent. This is exactly what one should expect. For with increased size of litter there would be fewer litters of 0 size, and the averages as actually found would more nearly represent the true averages.

Curves (Fig. 1) have been constructed showing the frequency of the litter sizes for certain of the types of matings. The data upon which they are based may be found in Table IX. A careful survey of these curves,

however, does not seem to lead to any very definite conclusions as to the special fitness of any of the various theories discussed in the preceding paragraphs. It will be seen that in the yellow \times yellow mating there are proportionately more litters of small size and fewer of large size than are to be found in the other matings. This is what one should theoretically expect on all the theories.

TABLE IX

LITTERS BORN DURING 1916-17, CLASSIFIED ACCORDING TO NUMBER OF YOUNG IN LITTER

No. in Litter	Number of Litters			
	Yel. ♀ \times Yel. ♂	Yel. ♀ \times Non-yel. ♂	Yel. ♂ \times Non-yel. ♀	Non.-yel. ♀ \times Non.-yel. ♂
1	1	1	1	
2	3	3	2	
3	21	5	1	1
4	25	11	3	4
5	25	13	4	7
6	24	18	9	5
7	25	11	6	12
8	10	10	12	3
9	3	11	4	4
10	3	3	5	5
11		2	3	1
Total...	140	88	50	42

Summing up, we may say that all of our evidence tends to confirm the conclusion of Castle and Little that in mice homozygous yellow zygotes are produced in the yellow \times yellow mating, but that these zygotes fail to develop normally after implantation in the uterus. Why this should be so is not evident and our investigation has not thrown any light on this point. It is possible a careful microscopical study of the embryos which die early might reveal some abnormality of development which would account for their failure to survive, but it is not probable that it would be of such a simple nature as the analogous cases of death of homozygous recessives lacking chlorophyll in corn and some other plants. It seems more probable that in mice there may be a "lethal factor," similar to those so well known in *Drosophila*, which is so closely linked to the factor for yellow that they are prac-

tically at the same locus and there is consequently no crossing over.

According to the investigations of Little (1915) an entirely similar condition obtains in regard to a dominant white spotting factor (*W*) in mice, which, like yellow, appears never to occur in a homozygous condition. Whether the homozygous individuals in this case also die at an early stage and might be found as dead embryos has not yet been determined. Little has, however, demonstrated (1917) that the two factors are independent in heredity, and that litters from yellows carrying *W*, mated *inter se*, average only three per litter (10 litters), while similar yellows mated to *ww* non-yellows have litters averaging 5 per litter (9 litters). Although the numbers are small the percentage relation for the two matings, 60 per cent., is quite close to the theoretical expectation, 56.3 per cent.

LITERATURE CITED

- Castle, W. E., and C. C. Little.
1910. On a Modified Mendelian Ratio among Yellow Mice. *Science*, N. S., Vol. 32, pp. 868-870.
- Cuénot, L.
1905. Les races pures et leurs combinaisons chez les souris. (4^{me} note.) *Arch. Zool. Expér. et Génér.*, 4^e Series, Vol. 3, Notes et Revue, pp. cxxiii-cxxvii.
- Cuénot, L.
1908. Sur quelques anomalies apparentes des proportions Mendéliennes. (6^e Note.) *Arch. Zool. Expér. et Génér.*, 4^e Series, Vol. 9, Notes et Revue, pp. vii-xv.
- Dunn, L. C.
1916. The Genetic Behavior of Mice of the Color Varieties "black-and-tan" and "red." *AMER. NAT.*, Vol. 50, pp. 664-675.
- Durham, F. M.
1911. Further Experiments on the Inheritance of Coat Colour in Mice. *Jour. Genetics*, Vol. 1, pp. 159-178. •
- Kirkham, W. B.
1917. Embryology of the Yellow Mouse. Proceedings of the Amer. Soc. of Zoologists, Abstracts. *The Anatomical Record*, Vol. 11, pp. 480-481.
- Little, C. C.
1915. The Inheritance of Black-eyed White Spotting in Mice. *AMER. NAT.*, Vol. 49, pp. 727-740.
- Little, C. C.
1917. The Relation of Yellow Coat Color and Black-eyed White Spotting of Mice in Inheritance. *Genetics*, Vol. 2, pp. 433-444.

SHORTER ARTICLES AND DISCUSSION

ON THE FAUNA OF GREAT SALT LAKE

IN a recent number of the *AMERICAN NATURALIST*¹ appeared a paper entitled "Notes on the Fauna of Great Salt Lake," by Dr. Chas. T. Vorhies. From observations made by the present writer in the region at the mouth of Bear River, Utah, during the summer and fall months from 1914 to 1916 inclusive, information is available that supplements in part the data given by Dr. Vorhies.

Bear River, the largest of the three main tributaries of Great Salt Lake, breaks up into a series of channels at its mouth and forms a great delta at the northern end of Bear River Bay. Immediately below the mouth of the river the waters of the bay are freshened by the incoming river water. Conditions here vary greatly however from day to day, and at present heavy salt water frequently comes up as far as Slaughter Island at the lower part of the marsh area in the river delta. Below this point the surface water coming from the river may be fresh while at a depth of a few inches a stratum of brine may overlie the mud. On calm days this overlapping proceeds for long distances. The prevailing summer winds however are from the south and southwest and these drive the salt water in toward the marsh nearly every afternoon.

It is common belief that the Southern Pacific cut-off has served as a dam to separate Bear River Bay from the main lake (cf. Vorhies, p. 494). For a considerable distance this causeway is made up of trestle work allowing free interchange of water from either side. Although tests of the density of the water were not made, the writer is certain that the difference in content of salts between the water on the north and south sides of the cut-off is slight while water sufficiently saline to enable the life characteristic of the Lake to flourish is found at least twelve miles above the cut-off and within four or five miles of the point at which the main channel of Bear River opens into what is known as South Bay.

¹ Vol. LI, No. 608, August, 1917, pp. 494-499.

Brine shrimp, *Artemia fertilis* Verrill, occurred at this point in enormous numbers and adults and larvæ of alkali flies, *Ephydra gracilis* Packard and *E. hians* Say, were abundant. *E. subopaca* Loew was less common. The brine shrimp were gathered in great masses, and took advantage of the slightest depressions in the mud as shelters against the ever-fluctuating currents. Thousands frequently gathered in the lee of the boat during periods of observation by the writer.

In May and June adult *Ephydra* were found on mud, laid bare by water receding from the high spring levels, in which alkalis were rising through surface evaporation. On these areas the flies formed dense masses several feet square. The insects were busily probing or kneading the mud with their proboscides so that the surface was heavily pitted or stippled with small depressions that were visible at a distance of several feet. The greater part of these insects were *Ephydra gracilis*.

The statement made by Dr. Vorhies (p. 498) that "enemies play no part in keeping down the numbers of *Artemia*, or of *Ephydra* in the larval stage" is not corroborated by observations of the present writer. After the first of September each year shovelers, *Spatula clypeata* (Linnaeus), began to congregate in the bay below the mouth of Bear River, and by October 1 thousands of these ducks were present. The birds lay in great banks on the open water, and it was not unusual to see such flocks that were at least two miles long and from one quarter to one half a mile broad. The shovelers were feeding almost entirely upon *Artemia fertilis* and larvæ and pupæ of *Ephydra*, and were crammed with them constantly. Usually this species of duck is not a good table bird but individuals shot here were all exceedingly fat, and the writer found them excellent eating. These ducks remained in fall until the fresh water bays were covered with ice. Another species of duck, the lesser scaup, *Marila affinis* (Eyton), came into this region from the north between October 2 and 12 each year, and by October 20, was abundant. These lesser scaups also frequented the lower bay, and, like the shovelers, fed to a large extent upon the brine shrimp and the immature stages of the alkali flies. At dusk on October 14, 1914, flocks of these ducks were observed from Promontory Point passing from Bear River Bay southwest past Fremont Island in the open lake. As there is no fresh water feeding ground in that direction it was assumed that they were going out

to feed at some favorable locality in the lake. Later in October the other ducks were joined by American goldeneyes, *Clangula c. americana* Bonaparte, while from observations it was certain that the green-winged teal, *Nettion carolinense* (Gmelin), at times fed upon this same food. The number of crustaceans and fly larvæ destroyed by these birds must be enormous.

In addition to these ducks great flocks containing thousands of Wilson's phalaropes, *Steganopus tricolor* Vieillot, and northern phalaropes, *Lobipes lobatus* (Linnæus), are found on the salt water during migrations, where these birds likewise feed upon the brine shrimp and the fly larvæ and pupæ. During October and November flocks of eared grebes, *Colymbus nigricollis californicus* (Heermann), were found on the lake along the cut-off where their food must have been taken from the same supply as none other suitable is found. It may be mentioned here that a considerable number of shovelers and many thousand eared grebes winter on Owen's Lake in California, where saline conditions are similar to those in Great Salt Lake, and where a similar fauna is found.

Avocets, *Recurvirostra americana* Gmelin, and black-necked stilts, *Himantopus mexicanus* (Müller), also fed upon *Artemia* and *Ephydra* at the mouth of Bear River, and no doubt these animals furnished food to other shore birds. Definite data on this point is not at hand, however, as all of the shore bird stomachs collected there have not yet been examined. It is probable that the ring-billed gull, *Larus delawarensis* Ord, and California gull, *Larus californicus* Lawrence, also take this same food at times.

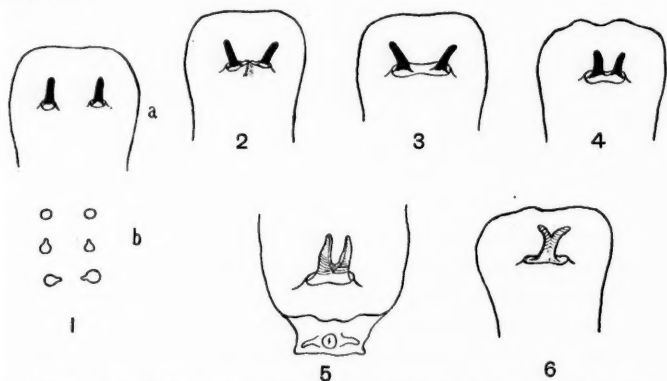
From the facts outlined above it will be seen that the toll taken by birds from the brine shrimp and alkali fly larvæ and pupæ during the course of a season constitutes a mass of individuals almost beyond comprehension. The digestion of food by the birds concerned is always a rapid process, and with soft-bodied animals like the brine shrimp a considerable mass would be consumed each day; and the same is true of the larvæ and pupæ of the alkali flies. The immense number of these creatures in the waters of the lake must be attributed to the large number of offspring produced rather than to an absence of enemies.

ALEXANDER WETMORE.

BIOLOGICAL SURVEY, WASHINGTON, D. C.

FUSION OF "RHINOPHORES" IN CHROMODORIS¹

THERE have been found during the present spring nine specimens of the nudibranch *Chromodoris zebra* Heilprin which form a series exhibiting an interesting gradation in the degree of coalescence of the "rhinophores." The animals were each of average adult size, 10-12 cm. in length. In none of these cases was there any evidence that the structural variations had resulted from injury. In the period over which these individuals were obtained there were also collected about 1,000 normal specimens of the same species. These figures give, however, no precise idea of the relative frequency of "rhinophore" variation, because a larger number of specimens had been collected in previous years without any occurrence of these variations being observed.



FIGS. 1-6. Outlines of anterior ends of *Chromodoris zebra* Heilprin, showing increasing degrees of fusion of the "rhinophores." Fig. 5, frontal view; the rest, dorsal aspects. Fig. 1a, the normal condition; Fig. 1b, variation in the edges of the "rhinophoral" collars of three individuals.

The bases of the two "rhinophores" of *C. zebra* are, as in other Dorids, normally surrounded by well developed individual cylindrical collars. The distal termination of a collar is usually circular in outline, but occasionally pointed at one side (Fig. 1). In two specimens the "rhinophoral" collars were closely approximated, after the fashion outlined in Fig. 2. Three specimens were found in which the "rhinophoral" collars, and the depressions into which the "rhinophores" are separately retracted when stimulated, had completely fused (Fig. 3). In

¹ Contributions from the Bermuda Biological Station for Research, No. 75.

these animals the two "rhinophores" themselves were separated by their normal distance of about 1 cm. The next step in "rhinophore" fusion is illustrated in Fig. 4, one example having been collected. In another specimen the "rhinophores" were found to be closely united at the base (Fig. 5), while in the remaining two specimens that exhibit fusion of the "rhinophores" (Fig. 6) the process of coalescence had been pushed much further, a single stalk, giving rise at its free end to two short diverging projections, representing the normal pair of "rhinophores."

When a "rhinophore" of *C. zebra* is locally stimulated by being touched, it is retracted within its pocket, the basal collar usually contracting over it, while the companion "rhinophore" on the other side of the animal is usually not contracted. In other words, the "rhinophores" are, with reference to their retraction, subject to independent bilateral control. The process of retracting the "rhinophore" consists of two phases—the "rhinophore" is itself contractile, and it is in addition pulled down into its pocket by the action of muscles situated at its base. With the fused "rhinophores," even in such cases as that illustrated in Fig. 6, the independent bilateral control of the organs is preserved. If one tip be stimulated, that side of the compound "rhinophore" is contracted, the other (unless the stimulation be severe) remaining inert. Under slightly stronger stimulation applied to one tip of a compound "rhinophore," the contraction of the organ itself is immediately followed by the traction of muscles upon the same side of the base of the double "rhinophore," resulting in a bending of the whole structure toward the point of excitation.

The reactions of the abnormal specimens therefore support the view that these abnormal "rhinophores" have been produced by a process of fusion, probably resulting from the original close approximation of "rhinophoral" Anlagen. Two cases have been available for experiment in which one of the normally placed "rhinophores" possessed a divided tip; these divided-tip "rhinophores," superficially not unlike the single median structure above described, gave no evidence of independent control for the two tips, both parts contracting together when one tip was irritated.

It would appear that the development of the collar surrounding the base of the "rhinophore" is directly dependent upon the growth of the latter structure; in every case there was a close

correspondence between the bulk of the "rhinophores" and the dimensions of the collar or collars.

W. J. CROZIER

AGAR'S ISLAND, BERMUDA

NOTE ON THE HABITAT OF GEONEMERTES AGRICOLA¹

THE terrestrial nemerteans include a small number of species, all belonging, apparently, to one genus, but widely scattered over the world. They occur conspicuously on islands, some of which are well removed from any large mainland. The origin of these land nemerteans is a matter of some interest, and several suggestions have been made relative to the manner of their evolution. One of these terrestrial nemerteans, *Geonemertes agricola* (W.-S.), was found at Bermuda by v. Willemoes-Suhm (1874). The anatomy of this species was subsequently described in detail by Coe (1904), who gave some attention, also, to the habits of the worm. These observers, as well as Verrill (1902), agree that *G. agricola* is to be found "only along the shores of mangrove swamps and on the adjacent hillsides" (Coe, p. 566). Coe found it "not only above high-water mark but also for some distance along a zone which is covered for a short time each day with sea water," but noted that the intertidal individuals were "as a rule smaller than those living in the soil which is a little above the reach of the tide, but in earth which is nearly saturated with salt water."

Standing bodies of fresh water are absent in Bermuda. Coe consequently held that this particular species, at least, represents a land nemertean which has almost certainly been derived directly from a marine ancestor, and not, as Montgomery (1895, p. 483) had argued for the generality of land nemerteans, from a fresh-water form.

During the past several years I have repeatedly encountered *G. agricola* in a type of habitat which is significantly different from that recorded for this nemertean by the observers just quoted. In the neighborhood of every large or small mangrove "creek" or swamp which I have examined, the worm has been found, in relatively considerable quantities, well below low-water mark even at spring tides. The species occurs in the localities

¹ Contributions from the Bermuda Biological Station for Research, No. 76.

listed by Coe, but is also common among the masses and sheets of matted sea weeds (*Laurentia*, *Valonia*, *Halimeda*, and associated plants) which cover the bottom of Fairyland Creek. Specimens were also obtained from under rocks situated a few feet beneath low-water level, in muddy bays bordered by mangroves, such as Tucker's Bay in Harrington Sound. The individuals collected in these places embraced white forms, some with a tinge of pink, others decidedly pink (as in the "pale" form figured by Coe, 1904, Pl. 1). They were 30-60 mm. in length, and some contained embryos.

In June and in July young *Geonemertes* were gotten among sea weeds in Fairyland Creek; these were 6-12 mm. in length. They were identified principally through the microscopic examination of the stylets and other organs. The stylets and stylet basis in these young specimens were of the juvenile type for this species, as figured by Coe (1904, Pl. 25, Figs. 21, 24, 25). These young specimens were in some cases pure white, in others tinged with "smoky brown." I found no pinkish specimens less than 30 mm. in length.

The observations upon the specimens of this species inhabiting salt water indicate, as Coe concluded from his study of the land-living individuals, that liberation of the young occurs in June and in July. My largest examples of *G. agricola* from the water were obtained in the spring.

Large specimens of *G. agricola* are negatively phototropic, the ocelli occupying the region of the body most sensitive toward light. They orient away from the light with diagrammatic precision. This response leads to their being found, during the day, under stones and about the roots of algæ.

It is hardly possible to credit the view that *G. agricola* has extended the variety of its habitats during the brief time since Coe's studies were made (1903); it is therefore necessary to believe that this species of nemertean is not only terrestrial in the proper sense, but truly marine as well. There seems no good ground upon which to distinguish and separate the individuals found respectively on land, in the intertidal zone, and definitely in the sea water. The terrestrial "variety" may then be regarded as having originated, perhaps not so very long ago, from the form which is undoubtedly marine—unless one is prepared to believe that, introduced as a terrestrial form, it has at some time secondarily taken to the sea after a protracted evo-

lution as a land animal. This case seems to have some resemblance to that of a grapsoid crab at Bermuda, *Sesarma ricordi* M.-Edw., of which a terrestrial variety has been described by Verrill (1908, p. 328). It is my impression that the larger marine specimens of *G. agricola* are less hardy, more easily caused to fragment by handling, than are those taken on land. This may, however, be merely a physiological consequence of differences in habitat, which could be exhibited within the life-history of a single individual. I have not been able to keep the salt water specimens alive after abruptly transferring them to damp earth. The young individuals, however, are quite hardy, and seem capable of enduring this treatment for several days at least.

These observations add further, and possibly final, weight to the argument that some, at least, of the land nemerteans have proceeded directly from ancestors inhabiting salt water.

W. J. CROZIER

AGAR'S ISLAND, BERMUDA

REFERENCES

- Coe, W. R.
1904. The Anatomy and Development of the Terrestrial Nemertean (*Geonemertes agricola*) of Bermuda. *Proc. Bost. Soc. Nat. Hist.*, Vol. 31, pp. 531-570, pl. 23-25. (Contrib. Bermuda Biol. Sta., No. 4.)
- Montgomery, T. H., Jr.
1895. The Derivation of the Freshwater and Land Nemerteans, and Allied Questions. *Jour. Morph.*, Vol. 11, pp. 479-484.
- Verrill, A. E.
1902. The Bermuda Islands. (Repr. from *Trans. Conn. Acad. Sci.*, Vol. 11, with changes.) New Haven, x + 548 pp., 38 pl., index.
1908. Decapod Crustacea of Bermuda. I. *Brachyura* and *Anomura*. Their Distribution, Variations and Habits. *Trans. Conn. Acad. Arts and Sci.*, Vol. 13, pp. 299-474, pl. 9-28.
- Willemoes-Suhm, R. Von
1874. On a Land Nemertean found in the Bermudas. *Ann. and Mag. Nat. Hist.*, ser. 4, Vol. 13, pp. 409-411, pl. 17.

NOTES AND LITERATURE

SUNSPOTS, CLIMATIC FACTORS AND PLANT ACTIVITIES

VARIATIONS in solar radiation, if of sufficient magnitude, should be followed by variations in terrestrial climatic conditions. In the absence of long series of determinations of the heat radiated by the sun, meteorologists have turned to variation in the number of sunspots as a possible factor underlying variation in the climatic factors. This involves the assumption that a period of many sunspots differs from a period of few spots in heat and light radiation.

If such climatic factors as heat and precipitation be closely related to the number of sunspots, the number of sunspots should be a factor of importance in determining plant activities. In recent years attempts have been made to correlate growth, especially that recorded in the annual rings of trees, with number of sunspots.¹

Since any attempt to relate growth phenomena to sunspot number presupposes relationships between climate and sunspot frequency, the botanist should be interested in the attempts of the meteorologist to ascertain the relationship between solar and terrestrial atmospheric phenomena. The purpose of this review is to call attention to certain recent discussions of this subject. For a review of earlier literature, the reader must refer to Hann's "Handbook of Climatology" and to the careful discussion from the biological side in Chapter XIX of Huntington's "Climatic Factor."²

Heretofore, those who have discussed the interdependence of terrestrial and solar phenomena have been content to plot curves for the two phenomena and to determine the existence of relationship between them by similar trends in the two curves.

The sources of error in such a method are very great. Furthermore, it gives no quantitative measure of intensity of relationship. Such a measure can only be secured by means of some correlation or contingency coefficient.

¹ Douglas, A. E., "A Method of Estimating Rainfall by the Growth of Trees." In Ellsworth Huntington's "The Climatic Factor," pp. 101-121.

² Huntington, E., "The Climatic Factor as Illustrated in Arid America," Pub. Carn. Inst. Wash., 192, 1914.

Walker,³ the director general of observatories for India, has made a great advance in the investigation of the possible relationship between the number of sunspots and meteorological phenomena by applying the modern methods of correlation to the problem.

I have tabulated his three series of correlations for a large number of stations widely distributed over the earth, with the results given in the accompanying table.

Intensity of Correlation	Frequency of Correlations		
	Sunspots and Rainfall	Sunspots and Temperature	Sunspots and Pressure
-.59 to -.53	—	2	—
-.52 to -.46	1	1	2
-.45 to -.39	1	6	2
-.38 to -.32	6	7	3
-.31 to -.25	9	10	8
-.24 to -.18	17	12	8
-.17 to -.11	15	15	8
-.10 to -.04	21	14	15
-.03 to +.03	27	16	11
+.04 to +.10	13	4	8
+.11 to +.17	18	5	10
+.18 to +.24	9	4	8
+.25 to +.31	7	1	6
+.32 to +.38	6	—	2
+.39 to +.45	—	—	—
+.46 to +.52	1	—	—
Total number of stations.	151	97	91

Remembering that correlation is measured on a scale of -1 to $+1$, this table shows at once that there is no uniformity for the globe as a whole in the correlations between number of sunspots and either of the climatic factors considered. Instead coefficients for some stations are positive while those for others are negative. Thus as far as the data available go, they indicate that in some regions rainfall, temperature or barometric pressure are higher in periods of larger numbers of sunspots, whereas in other regions they are lower. The magnitude of the coefficients is generally low.⁴ Over thirty per cent. of the constants

³ Walker, Gilbert T., "Correlation in Seasonal Variation of Weather," IV-VI. Mem. Ind. Met. Dep., 21: 10-12, 17-118, 3 world maps, 1915.

⁴ These correlations are based on such small samples that for their full interpretation the theory of the distribution of small correlations now being developed by "Student," Soper, Fisher, Young, Cave, Lee and Pearson must be considered. Nothing brought out by the work of these writers, which will be reviewed later, invalidates the general correctness of the conclusions reached here.

lie between $+.10$ and $-.10$. The probable errors of the constants are of about this magnitude.

The average values of the three sets of correlations are:

For sunspots and rainfall, 151 stations, $\bar{r} = -.0175$

For sunspots and temperature, 97 stations, $\bar{r} = -.1360$

For sunspots and pressure, 91 stations, $\bar{r} = -.0331$

While these averages are exceedingly small, all are negative in sign, indicating that for the globe as a whole lower rainfall, temperature and barometric pressure are associated with greater numbers of sunspots.

The same relationship is apparent if only the 76 stations for which all three relationships have been calculated be considered. The averages are:

For sunspots and rainfall, $\bar{r} = -.0349$

For sunspots and temperature, $\bar{r} = -.1534$

For sunspots and pressure, $\bar{r} = -.0486$

If in obtaining the average correlations the constants for the several stations are weighted with the number of years for which records are available the averages, indicated by the bars, are:

For sunspots and rainfall, 151 stations, $\bar{r} = -.0103$

For sunspots and temperature, 97 stations, $\bar{r} = -.1243$

For sunspots and pressure, 91 stations, $\bar{r} = -.0387$

Thus, however calculated, the averages indicate generally negative values of the correlation coefficient for all three relationships.

The same fact is brought out if the coefficients are classified according to sign only. Thus:

Number of Sunspots and:—	Frequency of Posi- tive Correlation	Frequency of Zero Correlation	Frequency of Neg- ative Correlation
Rainfall	70	1	80
Temperature	18	4	75
Pressure	38	3	50

For all three relationships the negative correlations are more numerous than those which are positive in sign.

In a brief review it is quite impossible to give in detail the meteorological considerations discussed in the original memoirs. Furthermore, the most of these do not directly concern the botanist. The conclusions of practical biological importance to be drawn from Walker's investigations seem to be the following:

a. The relationship between the number of sunspots and the annual record of terrestrial meteorological phenomena is very slender indeed. It is so slight that at the present time it is impossible to assert on the basis of the data of any one station alone that any relationship at all exists. Thus, as far as they go, these data hold out very little hope to the biologist of being able to correlate plant activities with sunspot number, unless light intensity be the means of solar influence.

b. For rainfall and barometric pressure the correlations are especially low. They average practically zero, but are apparently on the whole negative in sign.

c. The correlation between number of sunspots and terrestrial temperature is the most consistent and substantial of the three. The coefficients average about $-.14$. Thus years of larger numbers of sunspots are in the long run years of lower, *not higher*, terrestrial temperature.⁵

These results are directly opposed to the theories which seem to have prevailed among many writers.

J. ARTHUR HARRIS

⁵ Possibly, as Walker suggests, superheating in equatorial regions may raise the temperature in the upper air but lower that at ground level. The temperatures in which the botanist is primarily interested are, however, those which may influence the film of vegetation which covers the globe.

INDEX

THE NAMES OF CONTRIBUTORS ARE PRINTED IN SMALL CAPITALS.

- Alcohol and White Mice, L. B. NICE, 596
- ALLARD, H. A., Synchronism and Synchronic Rhythm, 438
- Allelomorphs, Multiple, and Modifying Factors in Relation to Selection, H. S. JENNINGS, 301; Reversible Transformability of, H. TERAQ, 690
- Amphibia, The Coal Measures, of North America, WM. K. GREGORY, 311
- Anatomical Literature, Sources of, ROY L. MOODIE, 193
- AWN, The Inheritance of the Weak, in *Avena* Crosses, H. H. LOVE and A. C. FRASER, 481
- BAUMBERGER, J. P., Solid Media for rearing *Drosophila*, 447
- Behavior, Synchronism and Synchronic Rhythm in, H. A. ALLARD, 438
- Biocharacters as Separable Units of Organic Structure, HENRY FAIRFIELD OSBORN, 449
- Biological, Facts and Bud-variation, E. M. EAST, 129; Significance of Animal Coloration, W. H. LONGLEY, 257; Enigmas and the Theory of Enzyme Action, LEONARD THOMPSON TROLAND, 321
- Biometric Studies on the Somatic and Genetic Physiology of the Sugar Beet, J. ARTHUR HARRIS, 507
- Blending and Mendelian Inheritance, A. C. and A. L. HAGEDOORN, 189
- BRIDGES, CALVIN B., the Variable Force Hypothesis of Crossing Over, 370
- Bud-variation, E. M. EAST, 129
- CASTLE, W. E., Piebald Rats and Multiple Factors, 102
- Chromodoris, Fusion of Rhinophores in, W. J. CROZIER, 756
- Chromosomes, Somatic, present in *Oenothera* Mutants and Hybrids, ANNE M. LUTZ, 375
- CLAUSEN, R. E. and T. H. GOOD-SPEED, Mendelian Factor Differences *versus* Reaction System Contrasts in Heredity, 31, 92
- COCKERELL, T. D. A., European Fossil Fish-scales, 61
- Coloration, Animal, W. H. LONGLEY, 257
- Control, Distributional, Field Tests of, JOSEPH GRINNELL, 115
- Correlation Formule and the Problems of Varietal Differences in Disease Resistance, J. ARTHUR HARRIS, 238
- Crosses, *Avena*, Inheritance of the Weak Awn in, H. H. LOVE and A. C. FRASER, 481; Wheat, Linked Quantitative Characters in, GEORGE F. FREEMAN, 683
- Crossing Over, the Variable Force Hypothesis of, CALVIN B. BRIDGES, 370
- Crown Gall, Resistance of *Prunus* to, CLAYTON O. SMITH, 47
- CROZIER, W. J., Migrations of Tropical Nudibranchs, 377; Multiplication by Fission in Holothurians, 560; Rhinophores in *Chromodoris*, 756; The Habitat of *Geonemertes agricola*, 758
- DANCHAKOFF, VERA, Differentiation in the Developing Organism, 419
- DAINES, L. L., The Flora of Great Salt Lake, 499
- DAVENPORT, CHARLES B., The Personality, Heredity and Work of Charles Otis Whitman, 5
- Dean and Eastman's Bibliography of Fishes, OLIVER P. HAY, 383
- Deer-mice, The Role of Isolation in the Formation of a Race of, F. B. SUMNER, 173
- Differentiation by Segregation and Environment in the Developing Organism, VERA DANCHAKOFF, 419
- Dimidium Nasutum, Mutation in, S. O. MAST, 351
- Distributional Control, JOSEPH GRINNELL, 115; Data, ELLIS L. MICHAEL, 572

- Drosophila*, Solid Media for rearing, J. P. BAUMBERGER, 447; *busckii* Coq, Mutations in, DON C. WARREN, 699
- DUNN, L. C., Nucleus and Cytoplasm as Vehicles of Heredity, 286
- EAST, E. M., The Bearing of Biological Facts on Bud-variation, 129
- Eastman and Dean's Bibliography of Fishes, OLIVER P. HAY, 383
- Ecology of the Protozoa, LEON AUGUSTUS HAUSMANN, 157
- Egg, Insect, The Genesis of the Organization of the, ROBERT W. HEGNER, 641, 705
- Enzyme Action and Biological Enigmas, LEONARD THOMPSON TROLAND, 321
- ERDMANN, RHODA, Sexual Process in the Myxosporidian Life Cycle, 719
- Evolution, from the Viewpoint of a Geneticist, A. FRANKLIN SHULL, 361; and Rats, A. C. and A. L. HAGEDOORN, 385
- "Factor," The Different Meanings of the, HOWARD B. FROST, 244
- Factors, Multiple, and Piebald Rats, W. E. CASTLE, 102; Modifying, and Multiple Allelomorphs and Selection, H. S. JENNINGS, 301
- Fertility, Inheritance of, in Sheep, EDWARD N. WENTWORTH and J. B. SWEET, 662
- Fish-scales, Fossil, European, T. D. A. COCKERELL, 61
- Fishes, The Migration of, DAVID STARR JORDAN, 186
- Fossil Fish-scales, European, T. D. A. COCKERELL, 61
- FRASER, A. C. and H. H. LOVE, The Inheritance of the Weak Awn in *Avena* Crosses, 481
- FREEMAN, GEORGE F., Linked Quantitative Characters in Wheat Crosses, 683
- Frequency, Mendelian Class, RAYMOND PEARL, 144
- FROST, HOWARD B., The Different Meanings of the Term "Factor" in Genetic Discussion, 244; Numbering Plants in Pedigree Cultures, 429
- GATES, R. R., The Mutation Theory and the Species Concept, 577
- Gene, The Theory of the, T. H. MORGAN, 513
- Genesis of the Organization of the Insect Egg, ROBERT W. HEGNER, 641, 705
- Genetics versus Paleontology, WM. K. GREGORY, 622
- Geonemertes agricola, Habitat of, W. J. CROZIER, 758
- Germ Plasm of *Enothera*, ROBERT T. HANCE, 567
- GOODSPEED, T. H. and R. E. CLAUSEN, Mendelian Factor Differences versus Reaction System Contrasts in Heredity, 31, 92
- Great Salt Lake, Fauna of, CHAS. T. VORHIES, 494; ALEXANDER WETMORE, 753; Flora of, L. L. DAINES, 499
- GREGORY, WM. K., The Coal Measures Amphibia of North America, 311; Genetics versus Paleontology, 622
- GRINNELL, JOSEPH, Field Tests of Theories concerning Distributional Control, 115
- HADLEY, PHILIP, The Case of *Trichomonas*, 209
- HAGEDOORN, A. C. and A. L., Blending and Mendelian Inheritance, 189; Rats and Evolution, 385
- HANCE, ROBERT T., The Germ Plasm of *Enothera*, 567
- HARRIS, J. ARTHUR, The Application of Correlation Formulae to the Problem of Varietal Differences in Disease Resistance, 238; Biometric Studies on the Somatic and Genetic Physiology of the Sugar Beet, 507; Sunspots, Climatic Factors and Plant Activities, 761
- HAUSMANN, LEON AUGUSTUS, The Ecology of the Protozoa, 157
- HAY, OLIVER P., Dean and Eastman's Bibliography of Fishes, 383
- HEGNER, ROBERT W., The Genesis of the Organization of the Insect Egg, 641, 705; Singing Mice, 704
- Heredity, Mendelian Factor Differences versus Reaction System Contrasts in, T. H. GOODSPEED and R. E. CLAUSEN, 31, 92; Nucleus and Cytoplasm as Vehicles of, L. C. DUNN, 286
- Holothurians, Multiplication by Fission in, W. J. CROZIER, 560
- IBSEN, HEMAN I. and EMIL STEIGLEDER, Death in Utero of the Homozygous Yellow Mouse, 740
- Inbreeding: Degrees of Kinship, RAYMOND PEARL, 545; A Single

- Numerical Measure of the Total Amount of, RAYMOND PEARL, 636
- Inheritance, Mendelian, and Blending, A. C. and A. L. HAGEDOORN, 189; of the Weak Awn, H. H. LOVE and A. C. FRASER, 481; of Fertility in Sheep, EDWARD N. WENTWORTH and J. B. SWEET, 662
- Isolation, The Rôle of, in the Formation of a Race of Deer-mice, F. B. SUMNER, 173
- JENNINGS, H. S., Modifying Factors and Multiple Allelomorphs and the Results of Selection, 301
- JONES, DONALD F., Linkage in *Lycopersicum*, 608
- JORDAN, DAVID STARR, The Migration of Fishes, 186
- Kinship, Degrees of, The Measurement and Numerical Expression of, RAYMOND PEARL, 545
- LINDSTROM, E. W., Linkage in Maize, 225
- Linkage in *Lycopersicum*, DONALD F. JONES, 608
- Linked Quantitative Characters in Wheat Crosses, GEORGE F. FREEMAN, 683
- LITTLE, C. C., Multiple Factors in Mice and Rats, 457
- LONGLEY, W. H., The Selection Problem, 250; The Biological Significance of Animal Coloration, 257
- LOVE, H. H. and A. C. FRASER, The Inheritance of the Weak Awn in Certain Avena Crosses, 481
- LUTZ, ANNE M., Characters Indicative of the Number of Somatic Chromosomes present in *Enothera* Mutants and Hybrids, 375
- MAST, S. O., Mutation in *Didinium Nasutum*, 351
- Mendelian, Factor Differences *versus* Reaction System Contrasts in Heredity, T. H. GOODSPEED and R. E. CLAUSEN, 31, 92; Class Frequency, RAYMOND PEARL, 144; SEWALL WRIGHT, 373; Inheritance and Blending, A. C. and A. L. HAGEDOORN, 189
- Mice, and Rats, Multiple Factors in, C. C. LITTLE, 457; White, and Alcohol, L. B. NICE, 596; Singing, ROBERT W. HEGNER, 704
- MICHAEL, ELLIS L., A Morphological Prediction from Distributional Data and its Verification, 572
- Migration, of Fishes, DAVID STARR JORDAN, 186; of Tropical Nudibranchs, W. J. CROZIER, 377
- MOODIE, ROY L., Sources of Anatomical Literature, 193
- MORGAN, T. H., The Theory of the Gene, 513
- Morphological Prediction from Distributional Data and its Verification, ELLIS L. MICHAEL, 572
- Multiple, Factors and Piebald Rats, W. E. CASTLE, 102; Allelomorphs and Modifying Factors and Selection, H. S. JENNINGS, 301; Factors in Mice and Rats, C. C. LITTLE, 457
- Mutation, A Wing, in *Piophila Casei*, THEOPHILUS S. PAINTER, 306; in *Didinium Nasutum*, S. O. MAST, 351; Theory and the Species Concept, R. R. GATES, 577; in *Drosophila busckii* Coq, DON C. WARREN, 699
- Mutants and Hybrids, *Enothera*, Number of Somatic Chromosomes present in, ANNE M. LUTZ, 375
- NICE, L. B., The Effects of Alcohol on White Mice, 596
- Notes and Literature, 311, 383, 507, 699, 761
- Nucleus and Cytoplasm as Vehicles of Heredity, L. C. DUNN, 286
- Nudibranchs, Tropical, Periodic Shoreward Migrations, W. J. CROZIER, 377
- Enothera*, Mutants and Hybrids, Number of Somatic Chromosomes present in, ANNE M. LUTZ, 375; An Attempt to modify the Germ Plasm through the Germinating Seed, ROBERT T. HANCE, 567
- Organism, Developing, Differentiation by Segregation and Environment in the, VERA DANCHAKOFF, 419
- Organization of the Insect Egg, Genesis of the, ROBERT W. HEGNER, 641, 705
- OSBORN, HENRY FAIRFIELD, Biocharacters as Separable Units of Organic Structure, 449
- PAINTER, THEOPHILUS S., A Wing Mutation of *Piophila casei*, 306
- Paleontology *versus* Genetics, WM. K. GREGORY, 622
- Panulirus argus*, Regeneration in, A. C. WALTON, 308
- PEARL, RAYMOND, The Selection Problem, 65; The Probable Error of a Mendelian Class Frequency, 144; Studies on Inbreed-

- ing: The Measurement and Numerical Expression of Degrees of Kinship, 545; A Single Numerical Measure of the Total Amount of Inbreeding, 636
- Pedigree Cultures, HOWARD B. FROST, 429
- (Peromyscus) The Role of Isolation in the Formation of a Race of Deer-mice, F. B. SUMNER, 173
- Piophilus casei, A Wing Mutation in, THEOPHILUS S. PAINTER, 306
- Protozoa, The Ecology of, LEON AUGUSTUS HAUSMANN, 157
- Prunus, Resistance to Crown Gall, CLAYTON O. SMITH, 47
- Quantitative Characters, Linked, in Wheat Crosses, GEORGE F. FREEMAN, 683
- Rats, Piebald, and Multiple Factors, W. E. CASTLE, 102; and Evolution, A. C. and A. L. HAGEDOORN, 385; and Mice, Multiple Factors in, C. C. LITTLE, 457
- Regeneration in Panulirus argus, A. C. WALTON, 308
- Resistance of Prunus to Crown Gall, CLAYTON O. SMITH, 47
- Selection, Problem, RAYMOND PEARL, 65; W. H. LONGLEY, 250; Modifying Factors and Multiple Allelomorphs, H. S. JENNINGS, 301
- Sexual Process in the Myxosporidian Life Cycle, RHODA ERDMANN, 719
- Sheep, Southdown, Inheritance of Fertility in, EDWARD N. WENTWORTH and J. B. SWEET, 662
- Shorter articles and discussion, 61, 102, 186, 238, 301, 370, 447, 494, 567, 636, 753
- SHULL, A. FRANKLIN, The Method of Evolution from the Viewpoint of a Geneticist, 361
- Singing Mice, ROBERT W. HEGNER, 704
- SMITH, CLAYTON O., Resistance of Prunus to Crown Gall, 47
- Species Concept and the Mutation Theory, R. R. GATES, 577
- STEIGLEDER, EMIL and HEMAN I. IBSEN, Death in Utero of the Homozygous Yellow Mouse, 740
- Sugar Beet, Biometric Studies on the Somatic and Genetic Physiology of the, J. ARTHUR HARRIS, 507
- SUMNER, F. B., The Role of Isolation in the Formation of a Narrowly Localized Race of Deer-mice (Peromyscus), 173
- Sun Spots, Climatic Factors and Plant Activities, J. ARTHUR HARRIS, 761
- SWEET, J. B. and EDWARD N. WENTWORTH, Inheritance of Fertility in Southdown Sheep, 662
- Synchronism and Synchronic Rhythm in Behavior, H. A. ALLARD, 438
- TERAO, H., On Reversible Transformability of Allelomorphs, 690
- Transformability, Reversible, of Allelomorphs, H. TERAQ, 690
- Trichomonas, The Case of, PHILIP HADLEY, 209
- TROLAND, LEONARD THOMPSON, Biological Enigmas and Enzyme Action, 321
- VORHIES, CHAS. T., The Fauna of Great Salt Lake, 494
- WALTON, A. C., Regeneration in Panulirus argus, 308
- WARREN, DON C., Mutations in Drosophila busckii Coq, 699
- WENTWORTH, EDWARD N. and J. B. SWEET, Inheritance of Fertility in Southdown Sheep, 662
- WETMORE, ALEXANDER, Fauna of Great Salt Lake, 753
- Whitman, Charles Otis, Personality, Heredity and Work of, CHARLES B. DAVENPORT, 5
- Wing Mutation in Piophilus casei, THEOPHILUS S. PAINTER, 306
- WRIGHT, SEWALL, The Probable Error of Mendelian Class Frequencies, 373
- Yellow Mouse, Homozygous, Death in Utero of the, HEMAN I. IBSEN and EMIL STEIGLEDER, 740

